PHENOTYPE

Issue 33 | Trinity Term 2019

NEUROSCIENCE & POETRY

Discovering the neuropoet in you!

PAGE 26

MICROBIOTA-GUT-BRAIN AXIS

Who's got the power?

PAGE 7

THE JOURNEY OF A FIRST-GENERATION GRADUATE

PAGE 24

"THE HARDEST THING I EVER DID WAS BEING AN ACADEMIC AND HAVING THREE YOUNG CHILDREN AT THE SAME TIME."

In conversation with the Vice Chancellor of Oxford Louise Richardson

PAGE 22

METABOLIC CONTROL OF SLEEP

PAGE 10

Your photo could be our next cover!
Turn to page 28 for details...

greiner
BIO-ONE

Cell Culture KEEPS ROCKING

when you have the right gear!

Samples available - contact us quoting 'Phenotype'

scan here

Tel: **01453 825255** email: **sales.uk@gbo.com** **www.gbo.com**

LETTER FROM THE EDITOR

Welcome to the HT19 issue.

It's been a real pleasure compiling this issue.

Turn to page 4 for an informative read on CRISPR and its journey from inception to the present state-narrated by Joey Riepsame, the Head of Genome Engineering at the Dunn School of Pathology.

On page 7, Maria Blanca Torraba elaborates on the emerging relationship between the microbiota in the gut and the brain. On page 10, Anissa Kempf vividly describes the impact of oxidative stress on sleep. On page 11, we have Jayanthiny writing about language learning and neuroplasticity, while on page 13 Shaked delves into the intricate relationship between inflammation and cell death.

We congratulate Robert Lees for winning last term's SNAPSHOT competition.

In our Science and Society section, we have two interesting, yet quite diverse articles written by Ines and Sunetra. On pages 14 and 15, Ines puts forth scientific arguments for her participation in the People's vote March, while on pages 16 and 17, Sunetra discusses how the concepts of Darwinism can shed light on Modern Capitalism.

On pages 19 and 20, Anna discusses effective science communication, while on pages 20 and 21, Heather talks about the future of lab automation.

Our latest "Aspire to Inspire' section features a very special interview with the Vice Chancellor of Oxford, Professor Louise Richardson. We thank her for such an open and candid conversation.

Accompanying this interview piece, we have Martine Abboud, the latest entrant to the Forbes 30 under 30 list, who speaks about her success mantra in academia and what propelled her life choices. Finally, we have Jayanthiny Kangatharan, describing her foray into neuropoetry and how it has such a strong connection to neuroscience.

The final pages of this issue feature a letter from Marina Kolesnichenko who together with her team founded Phenotype in 2008. In our previous issue we incorrectly named Sarah Iqbal as the founder. Sarah Iqbal served as the first editor.

We invite entries for our next issue, which will be a special double issue for MT19 and HT20. Deadline for all submissions is the 30th November, 2019. Please feel free to send us your pitches and ideas on oxphenotype@googlemail.com.

Follow us on twitter
@OxPhenotype
@Muliyilsonia

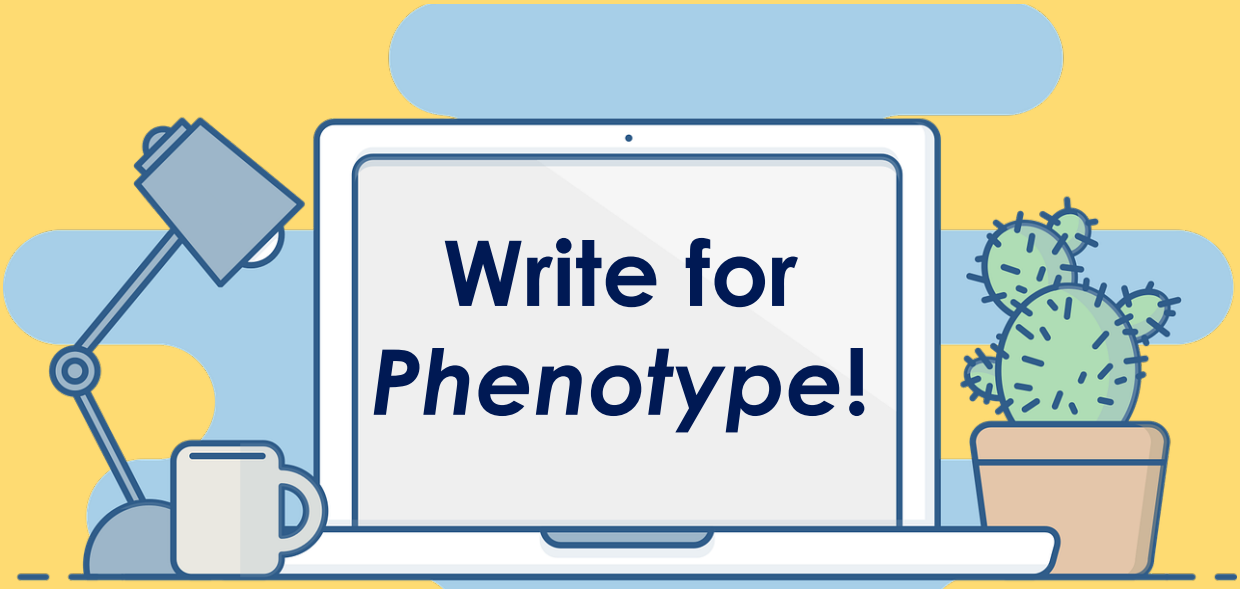
Sonia Muliyil

Editor-in-Chief

Phenotype is also available to read online via our website: **www.phenotypejournal.org**

Do you work at the very cutting edge of science?
Are you involved in exciting and influential outreach?
Are you a passionate writer?

Then this is the opportunity for you!



Visit Phenotype on our homepage
www.phenotypejournal.org and browse through a digital copy of this issue, join us on Facebook or follow **@OxPhenotype** on Twitter!

Write for any of our sections.

Submission deadline for the Special Double Issue is
30th November 2019

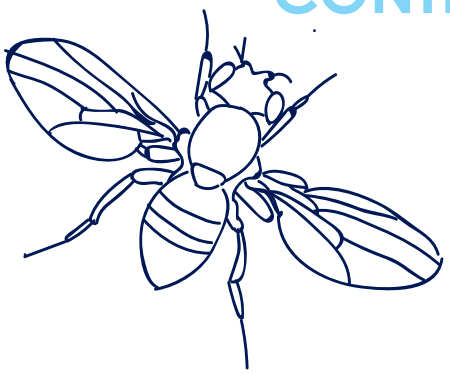
Get in touch!

Send pitches and submissions to
oxphenotype@googlemail.com

EDITORIAL TEAM

Issue 33 Trinity 2019

CONTENTS



FEATURES

SCIENCE & SOCIETY

ASPIRE TO INSPIRE

CTRL+X / CTRL+V

NEW DEVELOPMENTS IN CRISPR-BASED GENE EDITING

By Joey Riepsaame

Joey Riepsaame is Head of Genome Engineering Oxford (GEO) at the Sir William Dunn School of Pathology

The CRISPR gene editing tool made its debut in mainstream science in 2012 and has since revolutionized biological research and biotechnology in a similar fashion to the way PCR did back in the 1980s. In 2019, barely seven years after the seminal Science publication by Jinek *et al.*(1), CRISPR has made its way into the clinic; two cancer patients in Pennsylvania have recently been treated with autologous T cells harbouring CRISPR edited endogenous TCR and PD-1 loci. An additional 16 clinical trials are underway (including one at Imperial College, London), focusing on treating patients with different red blood cell malignancies such as β-thalassemia and sickle cell anaemia using CRISPR.

The tools used in the clinic involve the same two basic components as the CRISPR systems used in the lab; Cas9 and a single guide RNA (sgRNA). In brief, Cas9 is a programmable endonuclease derived from bacteria that can be loaded with a sgRNA to cut a very specific piece of DNA (protospacer) complementary to the 20nt spacer sequence located at the 5' end of the sgRNA (Figure 1). The only additional requirement for Cas9 to cut is that the genomic target needs to be flanked with a so-called protospacer-adjacent motif (PAM) that varies depending on the bacterial Cas9 species used. The most commonly used Cas9 nuclease, derived from *Streptococcus pyogenes*, recognizes a PAM sequence of NGG (where N can be any nucleotide) located on the non-target strand, directly downstream of the 20nt target sequence. Upon recognition of the correct PAM and target sequence, Cas9 cuts the DNA 3 nucleotides (nt) upstream of the PAM using its two endonuclease domains, RuvC and HNH. The resulting double-strand DNA break (DSB) is subsequently repaired by the error-prone non-homologous end joining (NHEJ) pathway, usually generating insertions or deletions (indels) of variable sizes. Alternatively, if a DNA repair template is provided, the cell can use the homology-directed repair (HDR) pathway to restore the lesion, thereby incorporating the desired mutation(s) present within the repair construct.

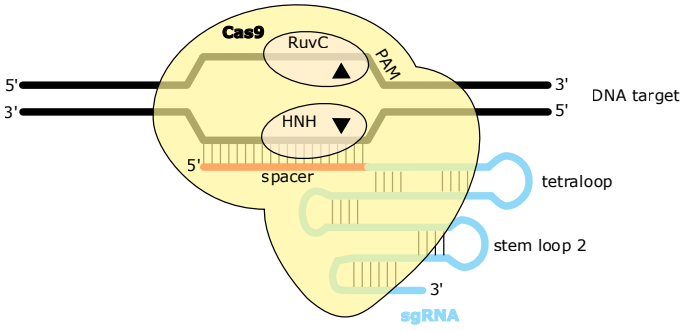


Figure 1. The RNA-guided S. pyogenes Cas9-sgRNA nuclease complex used for eukaryotic gene editing. Cas9 (yellow) loaded with a sgRNA (blue) bound to its cognate dsDNA target. Target recognition and cleavage (black triangles) by the HNH and RuvC endonuclease domains require protospacer sequence complementary to the spacer (red) and presence of the appropriate NGG PAM sequence at the 3' of the protospacer.

The first generation of CRISPR tools suffered from several issues, predominantly off-target effects (unintentional DNA changes at non-target sites), which prevented them from being used in the clinic. Fast forward six years and CRISPR is used in clinical trials all over the world.

Fast forward six years and CRISPR is used in clinical trials all over the world.

So, what has changed? Initial efforts to tackle the off-target effects problem included implementing double nicking using a Cas9 nickase variant (D10A) and truncating sgRNA protospacer sequences. Unfortunately, these options require a lot of tweaking, often lower on-target efficiency, and sometimes even an increase in off-target effects. At around the same time, several structural studies were published, providing a wealth of information about the way Cas9 interacts with the sgRNA and its cognate genomic DNA target. Based on these new insights, several groups managed to re-engineer Cas9 through a combination of rational design and directed evolution of various functional domains, resulting in new S. *pyogenes* (Sp) Cas9 variants with improved specificity and reduced off-target activity. Some of these include HyPaCas9

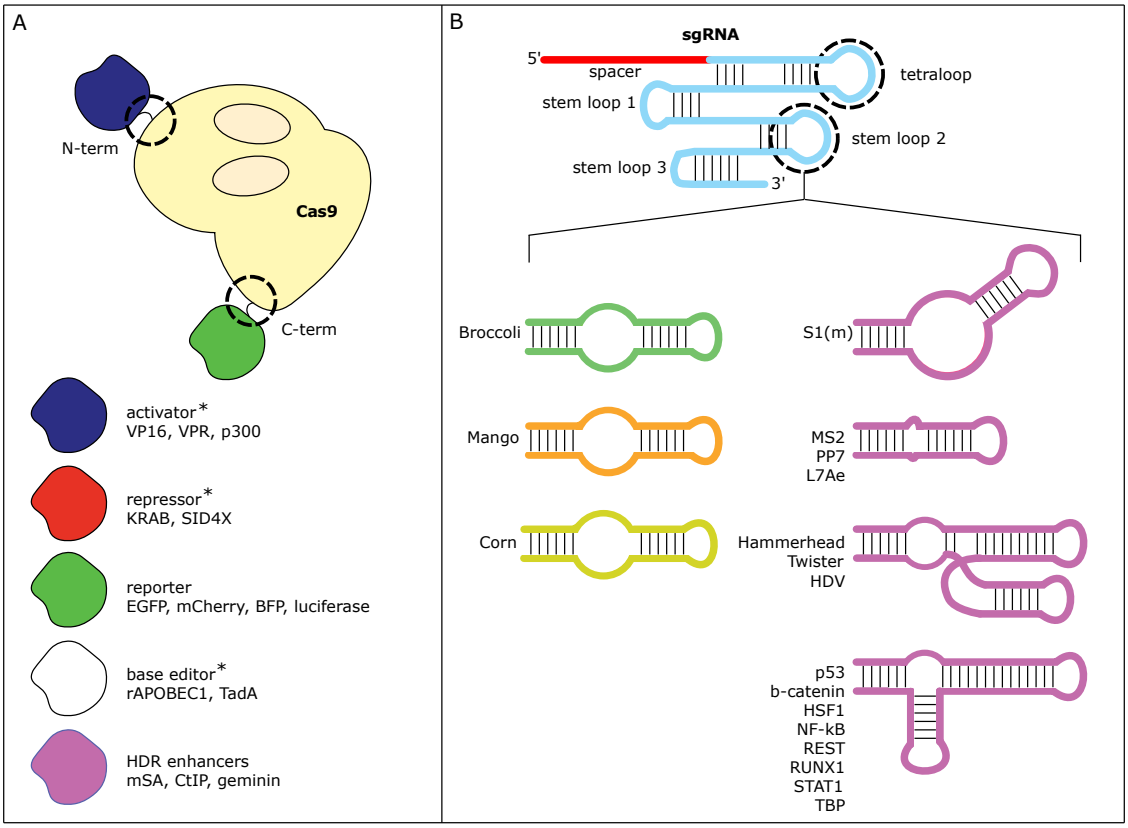


Figure 2. Expanding functionality of the S. pyogenes CRISPR system by Cas9 and/or sgRNA modifications. (A) Proteins can be targeted to almost any dsDNA sequence by fusing them to the N- or C-terminus (dashed circles) of Cas9. Examples include transcriptional activators, repressors, (fluorescent) reporters, base editors and HDR enhancers. Certain fusions (*) are exclusively used in combination with dCas9. (B) Functionality of the sgRNA can be expanded by incorporating functional RNA domains into the tetraloop and/or stem loop 2 (dashed circles). Examples include fluorescent aptamers (Broccoli, Mango, Corn), streptavidin-binding aptamers (S1(m)), RNA-binding protein scaffolds (MS2, PP7, L7Ae), self-cleaving ribozymes (Hammerhead, Twister, HDV) and protein-responsive aptamers (p53, β-catenin, HSF1 (heat shock factor 1), NF-κB (nuclear factor-kappaB), REST (RE1-silencing transcription factor) RUNX1 (runt-related transcription factor 1), STAT1 (signal transducers and activators of transcription), TBP (TATA-binding protein)) for conditional sgRNA activation.

(Doudna lab), eSpCas9 (Zhang lab), SpCas9-HF1 (Joung lab) and HiFi Cas9 (IDT). Interestingly, other groups have managed to develop the variants xCas9 (Liu lab) and SpCas9-NG (Nureki lab) which not only reduce off-target activity, but also have relaxed PAM restrictions, requiring only an NG sequence instead of NGG, thereby expanding the range of available target sites(2). Which of these newly developed Cas9 variants are going to be used in the first clinical trials is currently unknown, but HiFi Cas9 has been used successfully to correct the sickle cell disease (SCD)-causing p.E6V mutation in human CD34+ haematopoietic stem and progenitor cells (HSPCs) derived from patients with SCD(3).

Another way to reduce off-target effects is by reducing the amount of time Cas9 and sgRNAs are expressed in the target cell; the longer Cas9 is present in the cell, the bigger the chance of off-target effects. This can be achieved by changing the delivery method. Initially, plasmids encoding Cas9 and sgRNAs were introduced into the cell via lipofection or electroporation to edit a gene of interest. Alternatively, Cas9 can be delivered in the form of protein pre-loaded with an in vitro transcribed or chemically synthesized sgRNA(4). Once inside the cell, these Cas9 protein/sgRNA ribonucleoprotein (RNP) complexes edit their DNA target usually within an hour. Because of the relatively short half-life of Cas9, the RNP is inactivated within 24 hours, thereby limiting the chance of off-target effects. This "hit-and-run" approach is rapidly gaining popularity for *in vivo* (and *ex vivo*) gene editing. Another reason why plasmids are becoming less popular is because of the risk of random integration of all or part of the plasmid DNA into the host genome. Plasmid DNA can also be inserted in Cas9-induced on-target and off-target DSBs. These unwanted plasmid integrations are difficult to detect, especially at off-target sites, and often require elaborate and time-consuming assays, such as Southern Blotting and inverse/Splinkerette PCR. With RNP-based editing, there is no such risk. Whether RNPs or other delivery systems (such as non-integrating AAV viruses) will be used in the upcoming clinical trials is currently unknown.

Apart from these recent developments, there have been many other interesting studies describing various exciting new CRISPR gene editing tools. Although most of these are still in a pre-clinical stage, some are interesting to highlight. The CRISPR system is surprisingly amendable and tolerates a variety of modifications to its two basic components (Cas9 and sgRNA) without loss of functionality. Cas9 functionality can be further expanded by fusing different functional protein domains to either its N- or C-terminus (Figure 2A). An interesting example of this is Cas9-mSA, developed in Janet Rossant's lab, wherein monovalent avidin is fused to Cas9's C-terminus. This variant enables biotinylated DNA repair templates to be physically attached to the avidin moiety of the Cas9 fusion protein, resulting in vastly improved HDR efficiencies. A similar result can be obtained by C-terminal Cas9 fusions to the functional domains of proteins involved

in homologous recombination, such as CtIP and Geminin. Catalytically inactive (or "dead") Cas9 fusions are also often used, but not for gene editing purposes. Rather, they are implemented as molecular tools to direct proteins of interest to user-defined target DNA sequences. Examples include dCas9 fused to (fluorescent) reporter proteins to visualise DNA loci in vivo and dCas9 repressor fusions, which are often used as alternative for RNAi to transiently repress genes of interest through binding close to promoter sequences.

Base editors are the latest addition to the ever-expanding gene editing toolbox. These consist of dCas9 or Cas9 nickase, fused to a cytidine deaminase enzyme (rAPOBEC1) or RNA adenosine deaminase (TadA), which can convert DNA bases C•G to T•A or A•T to G•C, respectively. This allows users to generate point mutations within an approximately five-nucleotide window in the single-stranded DNA bubble created by Cas9. Base editing can achieve correction efficiencies of 15-75%, which is much higher than using conventional CRISPR/Cas9-mediated editing with HDR donors (typically 0.1-5%). Interestingly, base editors can convert DNA bases without introducing DSBs, due to the fact that rAPOBEC1 and TadA can only bind to single-stranded DNA, which is mediated by local denaturation of the target DNA upon dCas9:sgRNA binding. Therefore, indel formation is relatively low (0.1-5%) compared to conventional CRISPR/Cas9.

Cas9 crystal structure and sgRNA mutational studies have revealed that the so-called tetraloop and stem loop 2 of the sgRNA are largely dispensable for Cas9-mediated DNA cleaving. Therefore, extra layers of functionality can be added to the CRISPR system by incorporating functional RNA domains (ribozymes, aptamers, etc.) into the sgRNA scaffold (Figure 2B). New sgRNA scaffolds are much smaller and easier to clone than new Cas9 variants, so expanding functionality of the CRISPR toolbox through sgRNA alterations has become very popular recently and has already generated some interesting new tools. For example, several groups have managed to generate fluorescent sgRNAs by incorporating aptamers into the dispensable loops. Unlike chemically labelled sgRNAs (e.g. FITC, Cy3, etc.), these sgRNAs only become fluorescent when bound by Cas9 and exposed to certain cell-permeable small molecules. This can be useful for several types of kinetics studies, real-time imaging experiments or simply for assessing RNP transfection efficiencies.

Cas9 crystal structure and sgRNA mutational studies have revealed that the so-called tetraloop and stem loop 2 of the sgRNA are largely dispensable for Cas9-mediated DNA cleaving.

Another interesting development is sgRNAs with incorporated protein-binding aptamers. The best known example is the MS2 aptamer, capable of binding MS2 bacteriophage coat proteins. These aptamers have been around for a while and are widely used in genome-wide SAM CRISPRa (activation) library screens to recruit MS2-p65-HSF1 transcriptional activators to target gene promoter regions.

More recently, the streptavidin-binding aptamer S1(m) has been described as an alternative for Cas9-mSA to improve HDR efficiencies by physically anchoring streptavidin-bound biotinylated DNA repair templates to RNP complexes. Although current generation S1(m) sgRNAs have shown promising results, HDR efficiencies are still substantially lower compared to the Cas9-mSA system.

Finally, signal-responsive aptamers can be incorporated into the sgRNA scaffold to generate conditional CRISPR systems. In this approach, the sgRNA protospacer region is paired with an antisense stem so that it cannot bind to its target DNA. Binding of a specific ligand to the aptamer induces a conformational change that allows the guide region of sgRNA to interact with the corresponding DNA sequence to induce Cas9-mediated gene editing. Ligands include cell or tissue-specific signalling proteins, such as transcription factors RUNX1 and STAT1. Using a similar approach, Liu et al. (5) successfully achieved cell-specific gene activation by integrating NF-κB-responsive aptamers into the sgRNA scaffold. Since NF-κB is a major regulator of genes responsible for both the innate and adaptive immune response, these sgRNAs can be very useful in mediating conditional gene editing in cells exposed to inflammatory stimuli.

Seven years after its ground-breaking debut, CRISPR is still going strong. And with the first clinical trials on the way and an ever expanding CRISPR toolbox, it is unlikely CRISPR will be going away any time soon.

Seven years after its ground-breaking debut, CRISPR is still going strong.

To learn more about CRISPR and how it can help your research, feel free to contact the GEO facility at the Sir William Dunn School of Pathology or visit our website (https://www.path.ox.ac.uk/content/genome-engineering-oxford-geo) for additional information. We also offer a wide range of services to the Oxford scientific community, such as target strategy design, synthesis of in vitro transcribed sgRNAs (including guides with custom scaffolds or aptamers) and generation of custom gene-edited cell lines. For enquiries about our latest services, please send an email to joey.riepsaame@path.ox.ac.uk."

(1) Jinek M, *et al.* (2012) A Programmable Dual-RNA–Guided DNA Endonuclease in Adaptive Bacterial Immunity. *Science* 337(6096):816-821.
(2) Nishimasu H. *et al.* (2018) Engineered CRISPR-Cas9 nuclease with expanded targeting space. Science 361(6408):1259-1262.
(3) Vakulskas CA. *et al.* (2018). A high-fidelity Cas9 mutant delivered as a ribonucleoprotein complex enables efficient gene editing in human hematopoietic stem and progenitor cells. *Nat Med*. 24(8):1216-1224.
(4) Kim S. *et al.* (2014) Highly efficient RNA-guided genome editing in human cells via delivery of purified Cas9 ribonucleoproteins. *Genome Res.* 24(6):1012-1019.
(5) Liu *et al.* (2016) Directing cellular information flow via CRISPR signal conductors. Nat Methods. 13(11):938-944.

MICROBIOTA-GUT-BRAIN AXIS

WHO´S GOT THE POWER?

"Commensal microbiota have the ability to synthesise neurotransmitters and other active small molecules which can travel in the blood circulation to the brain."

By Maria Blanca Torroba

Maria Blanca Torroba is a Postdoctoral Researcher in Francis Szele's research group at the Department of Physiology, Anatomy and Genetics.

Have you ever heard that we carry more microorganisms (bacteria, fungi, etc.) in our body, particularly in our gut, than the amount of cells we are actually made of? And did you know that those microorganisms in our gut have direct access to the enteric nervous system, the so-called "second brain"? Can you then imagine the influence that these important microorganisms have in regulating how we function?

What if we are just skin-covered marionettes for microorganisms and they are the tyrants, bellowing orders from underground to clandestinely control humans? This may seem like a ridiculous notion, but perhaps by the end of the article, this may not seem like such a mad idea.

Evidence is accumulating to suggest that the microorganisms living in symbiosis with our host cells, collectively known as commensal microbiota, have some control over our body and mind. The most important population is the gut microbiota in the digestive tract, which accounts for over a kilogram of our body weight [1]. Estimated numbers vary across studies, but it is believed that there are at least as many microorganisms as host cells in our body. We are not alone!

The most important population is the gut microbiota in the digestive tract, which accounts for over a kilogram of our body weight.

Commensal microbiota have a very early influence in our lives, modulating embryonic and postnatal growth beyond what is encoded in our own genes. Those bacteria help shape the immune system, the neuroendocrine system and even the brain [1]. In daily life, those same microorganisms in the gut continue to have a role in gastrointestinal homeostasis.

The gut is a very interesting organ as it possesses its own nervous system, known as the enteric nervous system (ENS). The ENS is composed of approximately 500 million neurons and it can operate autonomously, using brain neurotransmitters such as GABA, dopamine or serotonin produced by the coexisting bacteria. Importantly, the microbiota-gut-ENS cooperation has roles beyond the digestive tract, affecting mood and higher cognitive functions as well [2]. This complex interaction is enclosed in the denomination "microbiota – gut – brain axis", which underpins my own *microorganism conspiracy theory*.

...the microbiota-gut-ENS cooperation has roles beyond the digestive tract, affecting mood and higher cognitive functions as well.

This axis has been extensively studied in germ-free (GF) mice which have no commensal bacteria whatsoever. These mice exhibit altered behavioural phenotypes relating to pain perception, learning and memory, mood and emotion. Notably, those alterations were significantly ameliorated by probiotic treatments, composed of live bacteria and yeast, promoted as having various health benefits [2]. In humans, equivalent bidirectional gut-brain correlations have been reported. Two strong examples are: a) the gut microbiota dysfunction repeatedly observed in depressed patients or b) the co-morbidity between irritable bowel syndrome (IBS) and the onset of depression [3].

But how could the gut microbiota affect the brain and our behaviour? Commensal microbiota have the ability to synthesise neurotransmitters and other active small molecules which can travel in the blood circulation to the brain. These molecules can modulate inflammation or activate neurons of the enteric nervous system. This information is then conducted to the

brain and back throughout the vagus nerve, as if it were a telephone line. This mechanism regulates immune activation, intestinal permeability, enteric reflex and neuro-endocrine signaling, the latter being dominated by the second major line of communication: the hypothalamic-pituitary-adrenal (HPA) axis. The HPA axis regulates the body´s reaction to stress, eventually triggering the systemic release of the "stress hormone", cortisol, from the adrenal glands, thus coordinating the response of different organs (including the brain) to a stressful situation. The gut microbiota, as the communication headquarters, regulates the information that circulates in these feedback loops and, therefore, how the brain responds (Figure 1).

The gut microbiota, as the communication headquarters, regulates the information that circulates in these feedback loops and, therefore, how the brain responds.

However, there are two sides to every story. In this one, the second side is that the survival, proliferation and wellbeing of the microbiota depend on the diet and the brain activity. Perhaps then, the microbiota-gut-brain axis functions more as a democracy where each player has a voice, rather than an absolute monarchy with some bacterial overlord playing a "game of cells" and controlling our minds.

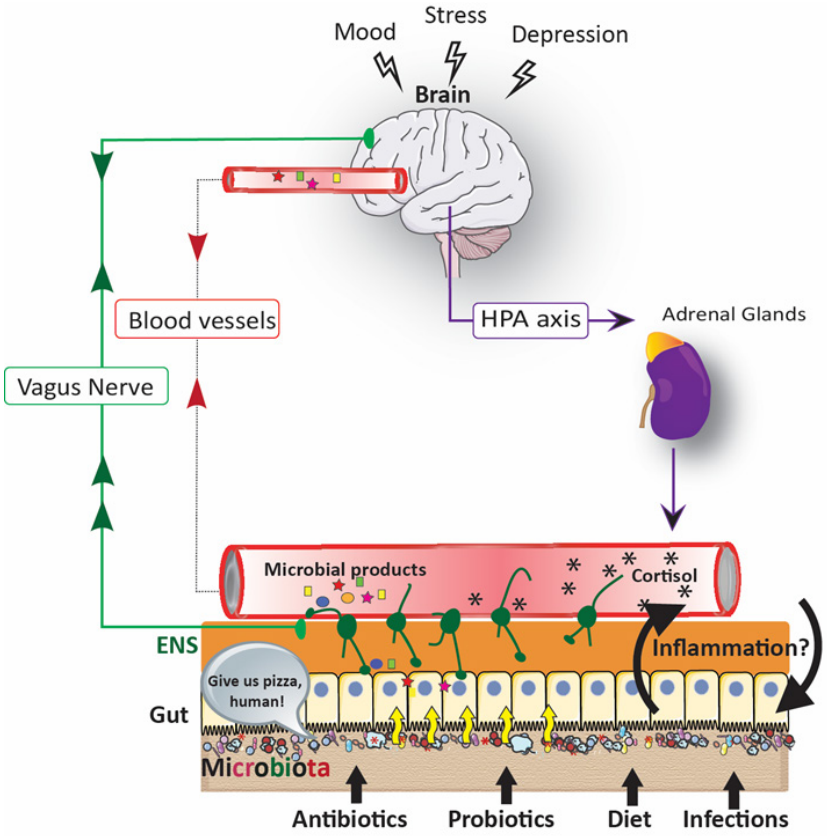
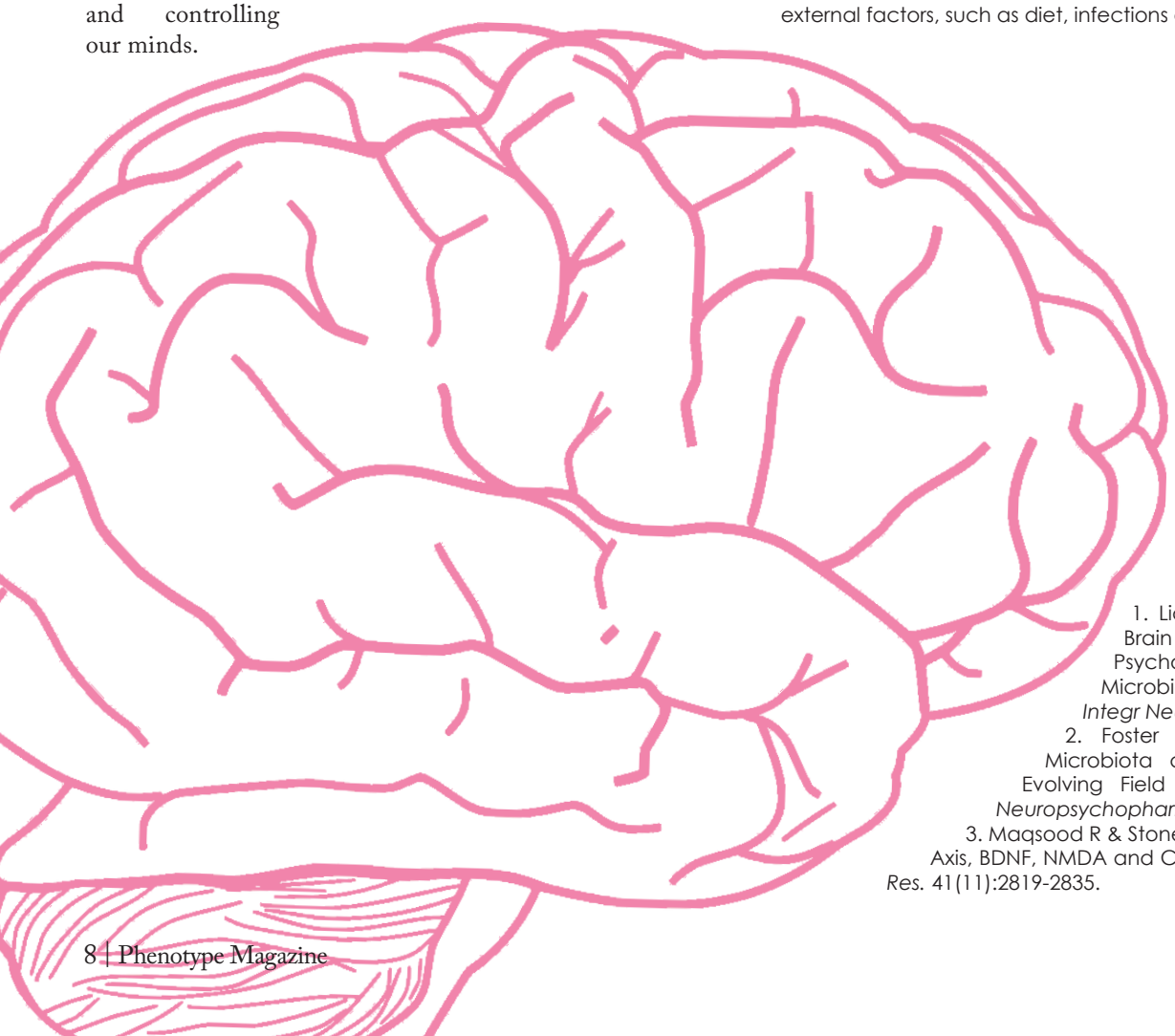


Figure 1. Microbiota – gut – brain axis. This scheme shows the bi-directional interactions between the microbiota in the gut, the enteric nervous system (ENS) and the brain throughout the vagus nerve, the circulating blood and the hypothalamus–pituitary–adrenal (HPA) axis. This network can be perturbed by many different external factors, such as diet, infections or stressful experiences.



1. Liang S *et al.*, (2018) Gut-Brain Psychology: Rethinking Psychology From the Microbiota-Gut-Brain Axis. *Front Integr Neurosci.* 11;12:33.

2. Foster JA *et al.*, (2016) Gut Microbiota and Brain Function: An Evolving Field in Neuroscience. *Int J Neuropsychopharmacol.* 19(5).

3. Maqsood R & Stone TW, (2016) The Gut-Brain Axis, BDNF, NMDA and CNS Disorders. *Neurochem Res.* 41(11):2819-2835.

SLEEP AND PHANTOM SOUNDS

Investigating tinnitus in the sleeping ferret.

By Linus Milinski

Linus Milinski is a DPhil student with Victoria Bajo, Vladyslav Vyazovskiy and Fernando Nodal in the Department of Physiology, Anatomy and Genetics.

A persistent noise; an ongoing hissing or ringing; a phantom sound without any source – this is what tinnitus can feel like. While these symptoms are often experienced during a common cold or after leaving a loud music concert, they are usually only temporary and therefore known as 'transient tinnitus'. Chronic tinnitus, on the other hand, is usually diagnosed if the phantom sound persists for extended periods of time, such as months or years. Especially the chronic condition is associated with stress, anxiety and sleep disturbances. Although an estimated 10-15% of people suffer from tinnitus in various degrees, we do not yet understand its cause or how it can be cured.

An estimated 10-15% of people suffer from tinnitus in various degrees, we do not yet understand its cause or how it can be cured.

Chronic tinnitus is commonly associated with hearing loss, which may be either age-dependent or caused by an insult to the auditory system, such as exposure to high-intensity noise (1). According to current understanding, a consequence of this hearing loss could be a persistent elevation of activity in some brain areas, leading to the experience of a phantom sound. However, it is unclear how brain activity actually changes when tinnitus develops. Furthermore, it is unknown whether these activity changes are restricted to the waking state, the sleeping state or occur during both.

Our research aims to elucidate the neuronal basis of chronic tinnitus, focusing on both the waking and sleeping state in order to obtain a fuller understanding. We hypothesise that there is a mechanistic connection between sleep and tinnitus. This is for two main reasons:

Firstly, as sleep is known to promote brain plasticity (2), we propose that sleep could be involved in the formation of chronic tinnitus by driving changes in the relevant brain networks (Fig. 1). Secondly, recent findings show that sleep can be locally regulated (3), meaning that one brain region can still be 'awake' while others are already 'asleep', depending on the sleep pressure or 'tiredness' of the respective regions. We propose that some regions might be continuously 'awake' in tinnitus. This would affect the arousal state of the brain and could disrupt natural sleep-wake dynamics (Fig. 1), fitting with the observation that tinnitus patients often report sleep problems.

We are investigating these hypotheses using the ferret as a model system. This allows us to monitor brain activity at both day and night for several months from before until after tinnitus has developed. We perform long-term recordings of brain activity by using implanted electrodes that detect electrical signals from single neurons and provide readouts for sleep and wakefulness. During the course of the experiment we induce tinnitus using noise over-exposure under sedation. In order to assess tinnitus, we use traditional hearing tests and also study performance in behavioural tasks known to be affected by the condition. These tasks are mainly based on detection of silent gaps embedded in sound (Fig. 2), as impairments in temporal processing of auditory signals are common in tinnitus. By doing this, we can correlate changes in brain activity with behavioural indicators of the condition. Finally, we are manipulating sleep dynamics in order to test their role in tinnitus development and expression.

This research will help us to understand the neuronal basis of chronic tinnitus and ultimately work towards new methods for its diagnosis and treatment. Indeed, by investigating sleep as a potential mediator for tinnitus, we are exploring a potential novel angle for future treatments.

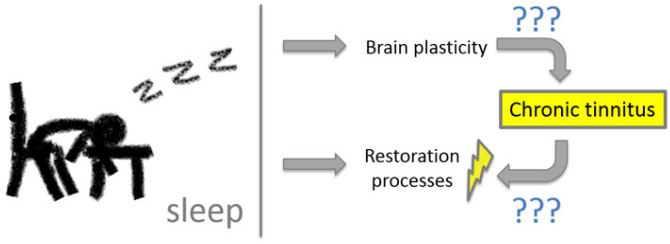


Figure 1: A possible connection between sleep and tinnitus. Top: Brain plasticity processes during sleep might be involved in tinnitus formation. Bottom: Aberrant brain activity associated with chronic tinnitus might disrupt normal sleep expression.

Figure 2: Gap detection protocol: The ability to discriminate between a continuous sound and a sound with an embedded silent gap can serve as an indicator for tinnitus: the phantom sound that is characteristic for tinnitus may fill the silent gap and impair successful discrimination.

1. Gold JR, Bajo VM (2014) Insult-induced adaptive plasticity of the auditory system. *Frontiers in Neuroscience*, 8, Article 110, 1-31
2. Tononi G, Cirelli C (2014) Sleep and the price of plasticity: from synaptic and cellular homeostasis to memory consolidation and integration. *Neuron*, 81(1), 12-34.
3. Vyazovskiy VV *et al.* (2011) Local sleep in awake rats. *Nature* 472, 443–447

METABOLIC CONTROL OF SLEEP

By Anissa Kempf

Anissa is a Post-doctoral fellow in the Meisenbock lab at the Centre for Neural circuits and behaviour

Sleep deprivation and sleep-related disorders greatly impact our society. Insufficient sleep adversely affects day-to-day performance and life quality, and costs billions per year to the UK economy (1). In spite of these detrimental effects, sleep deprivation treatment is inefficient and insufficient. Understanding the deeper mechanisms underlying its regulation, however, will be the best chance of substantial therapeutic improvement.

Two separate but interacting systems are thought to regulate sleep: the circadian clock and the sleep homeostat or somnostat. The circadian process synchronises sleep to the external 24-hour day-night cycle, and the homeostat senses internal changes to determine the amount and intensity of sleep needed, also referred to as sleep pressure. However, which mechanisms account for sleep pressure, as well as how sleep pressure signals induce sleep remain unknown. Sleep, and particularly why and how we sleep, will be better understood once we gain a molecular insight into the sleep homeostat.

Lesion studies in mammals have helped to identify a number of wake- and sleep-promoting nuclei within different brain regions, such as the hypothalamus, brainstem and basal forebrain/preoptic area. These nuclei were shown to mutually inhibit each other, giving rise to the 'flip-flop switch' model. In this model, only one of two brain states—asleep or awake—can be adopted at a time (2). Although the interpretation of this model is straightforward, these nuclei consist of very heterogeneous cell populations, hampering the cell type-specific dissection and the mechanistic understanding of the sleep homeostat in mammals.

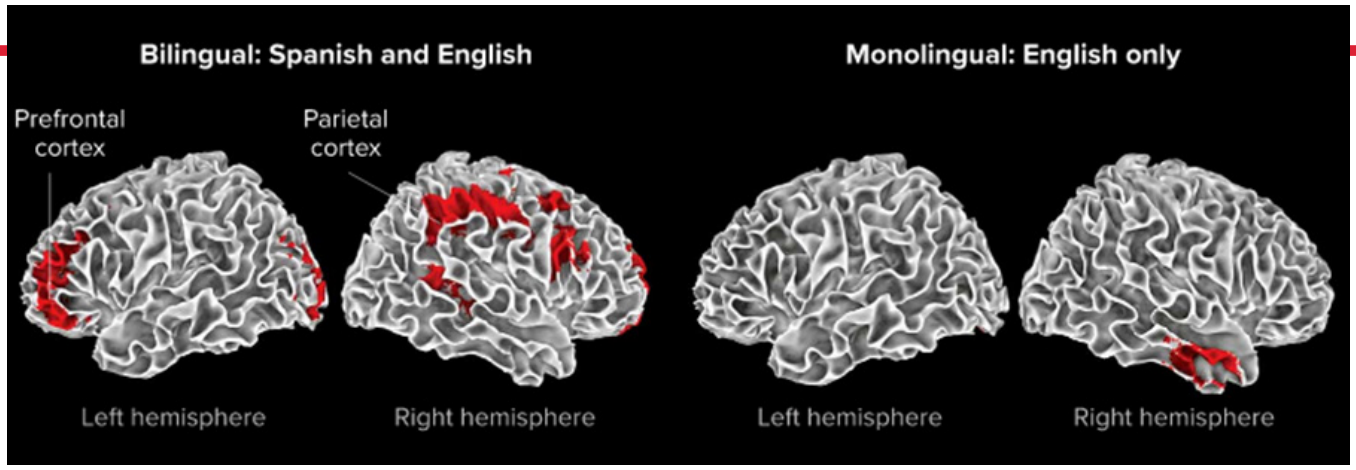
An analogous circuit controls sleep in the fruit fly *Drosophila melanogaster*, an animal model with fewer neurons and far less cell heterogeneity. Like mammals, fruit flies meet key behavioural criteria for the definition of sleep: reversible behavioural quiescence, reduced responsiveness to sensory stimulation, stereotypical postural changes and, most importantly, homeostatic regulation, expressed as an increase in sleep duration and intensity after prolonged periods of wakefulness (3). In the fruit fly, two-dozen cells projecting to the dorsal fan-shaped body (dFB) have been shown to act as bona fide sleep-control neurons. They promote sleep upon artificial activation, while their inhibition by wake-promoting dopaminergic neurons prevents sleep, reminiscent of the 'flip-flop' switch model (4, 5). Like mammalian sleep-active neurons, dFB neurons are GABAergic and peptidergic (6) and switch between two distinct electrophysiological states that strongly correlate with the wake/sleep state of the fly. They are electrically active (ON) when the fly is asleep, and silent (OFF) when the fly is awake. Most importantly, dFB neurons respond to homeostatic sleep pressure: they become more active after prolonged periods of wakefulness, e.g., after sleep deprivation, suggesting that sleep pressure is directly linked to the extent of their electrical excitability (7).

The excitability switch is, in turn, regulated by two antagonistic potassium currents conducted by the two-pore channel Sandman and the voltage-gated channel Shaker. While Sandman dominates during waking and imposes electrical silence, Shaker supports the activation and the tonic firing of dFB neurons during sleep. Therefore, the identification of the sleep-promoting signals that modulate Shaker activity will solve parts of the somnostat puzzle. We have recently shown that Shaker activation, and thus sleep, depends on the production of mitochondrial reactive oxygen species (ROS) (8). In order to produce the energy-carrying molecule ATP, cells rely on redox reactions carried out by a series of protein complexes in the electron transport chain. Depending on the metabolic state of the cell, these reactions can lead to the production of ROS. During waking (i.e., when sleep-promoting neurons are OFF and their energy demand is low) oxidative by-products of mitochondrial electron transport accumulate, modulate Shaker, and, as a consequence, activate dFB neurons promoting sleep. These findings are fascinating because they mechanistically link energy metabolism, oxidative stress and sleep, three processes independently implicated in ageing and neurodegenerative diseases. However, many mechanistic pieces remain to be investigated. For example, it is unclear whether the accumulation of ROS is the result of a cell-autonomous process and/or is triggered by external signals. Moreover, while the dFB neurons represent the output arm of the somnostat, other regions of the brain are also involved in sleep regulation (3). Future challenges will therefore be to address how different parts of the sleep circuitry are interconnected and induce sleep at a global level.

1. M. Hafner, M. Stepanek, J. Taylor, W. M. Troxel, C. van Stolk, Why Sleep Matters-The Economic Costs of Insufficient Sleep: A Cross-Country Comparative Analysis. *Rand Health* Q. 6, 11 (2017).
2. C. B. Saper, P. M. Fuller, N. P. Pedersen, J. Lu, T. E. Scammell, Sleep state switching. *Neuron*. 68, 1023–1042 (2010).
3. G. Artiushin, A. Sehgal, The Drosophila circuitry of sleep-wake regulation. Curr. Opin. *Neurobiol*. 44, 243–250 (2017).
4. J. M. Donlea, M. S. Thimgan, Y. Suzuki, L. Gottschalk, P. J. Shaw, Inducing sleep by remote control facilitates memory consolidation in Drosophila. *Science*. 332, 1571–1576 (2011).
5. D. Pimentel *et al.*, Operation of a homeostatic sleep switch. Nature. 536, 333–337 (2016).
6. J. M. Donlea *et al.*, Recurrent Circuitry for Balancing Sleep Need and Sleep. *Neuron*. 97, 378–389.e4 (2018).
7. J. M. Donlea, D. Pimentel, G. Miesenböck, Neuronal machinery of sleep homeostasis in Drosophila. *Neuron*. 81, 860–872 (2014).
8. A. Kempf, S. M. Song, C. B. Talbot, G. Miesenböck, A potassium channel β-subunit couples mitochondrial electron transport to sleep. *Nature*. 568, 230–234 (2019).

LANGUAGE LEARNING AND NEUROPLASTICITY

The effects of multilingualism on brain structure and function



Gray matter volume in bilinguals versus monolinguals: a study measured gray matter volumes in monlinguals and bilinguals. Red areas indicate where gray matter volumes were greater in Spanish and English bilinguals when compared to English monolinguals. These areas are the bilateral frontal and right parietal regions (8).

By Jayanthiny Kangatharan

Jayanthiny Kangatharan, PhD, is a Postdoctoral Research Assistant, Harvard University

Advanced understanding of bilingual language acquisition

During the first half of the 20th century speaking a foreign language was strongly discouraged as conventional wisdom considered learning a second language detrimental to cognitive development. It was only after 1962 when an extensive study by Peal and Lambert (1) on the effects of bilingualism on intellectual functioning was published that a positive image of bilingualism emerged: By comparing monolingual French speaking with English-French bilingual children, Pearl and Lambert found bilingual children to perform significantly better than monolinguals on verbal and non-verbal intelligence tests. They portrayed those bilinguals as someone with "[…] mental flexibility, a superiority in concept formation, a more diversified set of mental abilities." Subsequent research was able to support this suggested finding of general cognitive advantages in bilingualism that are not restricted to linguistic processing such as task-switching (2), and conflict monitoring (3).

The effects of acquiring more than one language on the structure and function of the brain

What are the implications of using more than one language on brain structure and function? Intending to speak one language over another means that the brain has to detect a conflict between languages, and resolve it by inhibiting responses from the unintended language, and selecting those responses from the intended language. A cortico-subcortical network is involved in the neural basis of the cognitive capacities that help bilinguals and multilinguals regulate the use of the intended language, while controlling for possible interferences from the unused but active language(s). This network coordinates the intricate process of language control and overlaps with neural structures underpinning domain-general executive control (4). Executive control includes a group of cognitive processes such as inhibitory control that are required to control behaviour to achieve goals that one has set for oneself. Neural network brain areas that become active outside of the language network are: the dorsolateral prefrontal cortex (DLPFC), the bilateral caudate, the cerebellum, the supplementary motor area, and the anterior cingulate cortex (ACC). The ACC is especially involved in monitoring and suppressing conflicting input in both linguistic and non-linguistic domains (5).

A study that used executive control tasks to compare conflict resolution abilities between multilinguals and monolingual speakers, revealed a lower activation in the ACC of multilinguals, which was correlated with greater local grey matter volume in that region. (6). This suggests that the ACC might be specially attuned to simultaneously manage multiple languages. Similarly, larger gray matter density was found in multilinguals than bilinguals within the posterior supramarginal gyrus (SMG), which connects two aspects of lexical knowledge: an anterior parietal area for processing phonology, and the angular gyrus for processing meaning (7). This finding correlates with a larger lexicon being used in multilinguals. Similarly, compared to monolinguals, bilinguals were reported to have increased gray matter densities in that region.

Gray matter volume in bilinguals versus monolinguals: a study measured gray matter volumes in monlinguals and bilinguals. Red areas indicate where gray matter volumes were greater in Spanish and English bilinguals when compared to English monolinguals. These areas are the bilateral frontal and right parietal regions (8).

The caudate nuclei represent a brain structure that has been involved in language control. They have been shown to be larger in bilinguals than in monolinguals (9). More recently, increased multilingual expertise was observed to correlate with bilateral caudate volume and also with regionally specific morphological changes of the left caudate nucleus (10).

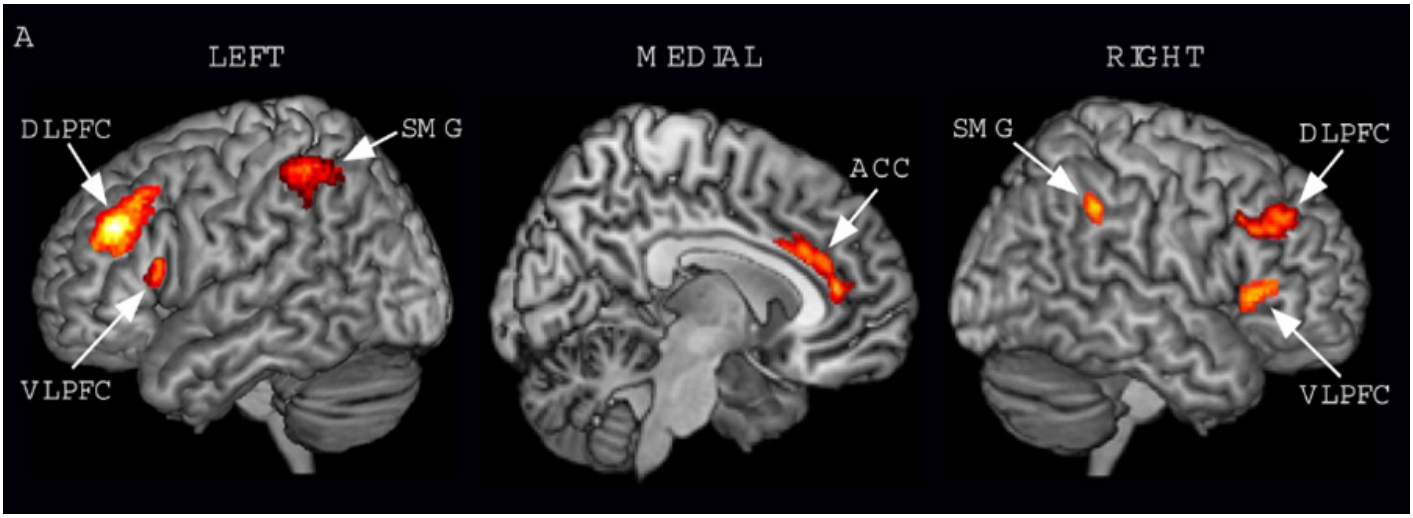
Relative to monolinguals, bilinguals have also been shown to engage a more distributed network of brain areas during language control tasks (11). Specifically, the observed larger frontal white matter, and higher measures of white matter microstructure are considered to allow lifelong bilinguals to use a larger network of brain regions.

Challenges and long-term consequences associated with multilingualism

Are there any challenges linked to the ability of speaking more than one language? It is thought that, despite its many advantages, managing more than one language will come at some cost. For example, the constant activity of two languages and the associated additional processing cost have been suggested to hinder bilinguals' skills in verbal communication compared to monolinguals of the same language (12).

Nevertheless, when one looks at the long-term consequences of bilingualism and multilingualism, it becomes evident that the many benefits are worth the cost. The bilingual advantage, for example, has been shown in older bilinguals who revealed better abilities in switching between tasks, resolving cognitive alternatives, and neglecting irrelevant information (13).

Such cognitive benefits are considered to have permanent and profound effects on brain health, as evidenced by research indicating that the ability to speak a second language leads to a larger cognitive reserve, whereby the onset of neurodegenerative diseases such as Alzheimer's disease is delayed. In the case of pathology and stress, it therefore appears as if the skills that one develops as a function of managing two or multiple languages can protect the brain against cognitive decline in later age and allow for functional cognition to operate for longer (15).



Compared to the non-switch condition, the switch condition leads to the noticeable activation of bilateral DLPFC, ventrolateral prefrontal cortex (VLPFC), ACC and bilateral SMG across all language speakers. However, older lifelong bilingual adults showed more efficient switching than older monolingual adults. Specifically, it was shown that older bilinguals had lower switch costs than older adult monolinguals within the left DLPFC, left VLPFC and ACC. (14).

1. Peal, E., & Lambert, W. (1962). The Relation of Bilingualism to Intelligence. Washington, DC: American Psychological Association.
2. Prior, A., & Gollan, H. (2011). Good language-switchers are good task-switchers: evidence from Spanish-English and Mandarin-English bilinguals. J. *Int. Neuropsychol. Soc*., 17, 682–691.
3. Costa, A., Hernández, M., Costa-Faidella, J., & Sebastián-Gallés, N. (2009). On the bilingual advantage in conflict processing: now you see it, now you don't. *Cognition*, 113, 135–149.
4. Abutalebi, J. & Green, D. (2007) Bilingual language production: the neurocognition of language representation and control J *Neurolinguistics*, 20: 242–275.
5. Pliatsikas, C. & Luk, G. (2016). Executive control in bilinguals: a concise review on fMRI studies. *Biling*. (Camb. Engl.), 53, 1689–1699.
6. Abutalebi, J., DellaRosa, P. & Ding, G. *et al.* (2013). Language proficiency modulates the engagement of cognitive control areas in multilinguals. *Cortex*, 49, 905–911.
7. Lee, H., Devlin, J. & Shakeshaft, C. *et al.* (2007). Anatomical traces of vocabulary acquisition in the adolescent brain. J. *Neurosci*., 27, 1184–1189
8. Olulade, O., Jamal, N., Koo, D. , Perfetti, C., LaSasso, C, & Eden, G. (2015). Neuroanatomical Evidence in Support of the Bilingual Theory. *Cerebral Cortex*, 26, 3196-3204.
9. Burgaleta M., Sanjuan A., Ventura-Campos N., Sebastian-Galles N, & Avila, C. (2016) Bilingualism at the core of the brain. Structural differences between bilinguals and monolinguals revealed by subcortical shape analysis. *Neuroimage*, 125, 437–445.
10. Hervais-Adelman, A., Egorova, N. & Golestani, N. (2018). Beyond bilingualism: multilingual experience correlates with caudate volume. *Brain Structure and Function*, 223, 3495-3502.
11. Olsen, K., Pangelinan, M., Bogulsk C., Chakravarty, M., Luk G., Grady, C., & Bialystok, E. (2015). "The Effect of Lifelong Bilingualism on Regional Grey and White Matter Volume." *Brain Research,* 1612, 128–139.
12. Bialystok, E., Craik, M., & Luk, G. (2012). Bilingualism: consequences for mind and brain. *Trends Cogn. Sci*.,16, 240–50
13. Bialystok, E., Craik, M., Green D., & Gollan T. (2009). Bilingual minds. *Psychol. Sci. Public Interest*, 10, 89–129.
14. Gold, B., Kim, C., Johnson, N., Kryscio, R., & Smith, C. (2013). Lifelong Bilingualism Maintain Neural Efficiency for Cognitive Control in Aging. *Journal of Neuroscience*, 33, 387-396.
15. Perquin, M., Vaillant, M., Schuller, M., Pastore, J., Dartigues, J., Lair, M., & Diederich, N. (2013). Lifelong exposure to multilingualism: new evidence to support cognitive reserve hypothesis. *PLoS One,* 8, e62030.

A MATTER OF LIFE AND DEATH

Inflammation and Cell Death

By Shaked Ashkenazi

Shaked Ashkenazi is a postdoctoral fellow in the lab of Matthew Freeman in Sir William Dunn School of Pathology. She is a research fellow of the Newton International Fellowship by the Royal Society and a Junior Research Fellow in Wolfson College.

Ancient Roman physicians described inflammation as a process of pain, swelling, heat and redness. For centuries, the molecular and cellular mechanisms of inflammation remained far from understood. Rudolf Virchow, the father of modern pathology, used a microscope to detect leukocytes in sites of inflammation. However, he completely misinterpreted their role in the disease. While he was right to realise that the white blood cells were involved in inflammatory symptoms and tissue damage, he failed to see that they were there to resolve the insult, rather than cause it. Despite his invaluable contribution to the field of molecular medicine, with regards to inflammation, Virchow was simply wrong. It was Ilya Metchnikoff who discovered that the true role of leukocytes in inflammation is to aid clearance of infections, and that damage to surrounding tissues is a part of the process.

In the late 19th century, surgeon William Coley noticed spontaneous remission occurring in cancer patients who experienced infections. He then tried to recapitulate the effect by injecting cancer patients with "Coley's Toxins", heat-inactivated bacteria, to induce the desired anti-tumour response, without risking fatal infections. This strategy had a moderate effect; however, it did not become a common practice, due to the development of more effective treatments. Nevertheless, this was one of the first pieces of evidence linking cancer with the immune system response to infections.

The most famous link between these two phenomena is the cytokine Tumour Necrosis Factor Alpha (TNFα). Although commonly known as a potent inducer of inflammation, its name suggests a significant role in a different cellular pathway, cell death. This protein plays a major part in orchestrating both of these arms. One arm is responsible for initiating inflammation, the desired immune response to invading pathogens. The other arm controls crucial cell death versus cell survival decisions, primarily by regulating apoptosis. Although the initial observation implied that TNFα is a pro-apoptotic agent, many further experiments described it as an anti-apoptotic factor, or in other words, pro-survival. This duality is not uncommon in cell biology. To this end, it is still unknown exactly how the cell death / survival outcome is determined.

The observations of chronic inflammation in the microenvironment of tumours, as well as the documented effects of inflammatory mediators on cancer progression, have led to inflammation being declared as the 7th hallmark of cancer. Therefore, it is not surprising that TNFα is involved in tumour progression, whether it is by promoting angiogenesis, enhancing immune evasion of cancer cells, or even simply by increasing cell survival. While Coley was right to suspect a connection between inflammation and tumours, it appears that the common link actually involves inflammation as a promotor of cancer, rather than a suppressor.

Although the dual role of TNFα is the best characterised example of the crosstalk between inflammation and cell death, it is not the only one. Recent discoveries suggest that autophagy is another form of cell death linked to inflammation and the response to infection. Autophagy is the process by which cellular components are specifically targeted, engulfed by a membrane and fused with lysosomes resulting in degradation. It is not surprising that such an efficient process could also be utilised to eliminate pathogens that successfully invade cells. One can easily observe the similarities between phagocytosis of extracellular pathogens, and autophagy of those that managed to penetrate the cells. Indeed, when the designated intracellular receptors detect an invading pathogen, two responses are initiated in parallel: the first is the assembly of the autophagy complex to target the pathogen for lysosomal degradation. The second is the initiation of an inflammatory signalling cascade, aimed at recruiting leukocytes to the site of infection.

Accumulating evidence now suggests that some chronic inflammation syndromes such as some cases of inflammatory bowel disease, result from genetic mutations in components of the autophagy-inflammation axis,

It is easy to see the evolutionary rationale behind cells utilising a single autophagy machinery for multiple stress responses, including infection. However, it is harder to rationalise why a different form of cell death, apoptosis (and to some extent necroptosis), should be so intimately linked with the response to infections. Homologues of TNFα and its receptor first appeared in corals and have been highly evolutionarily conserved since. Naturally, the immune system of these marine invertebrates is far inferior to that of mammals. It is possible that the dual role of TNFα is a form of "altruism", resulting from lack of a better mechanisms to fight infections. Accordingly, in the absence of direct means to eliminate a pathogen, an infected cell should kill itself to prevent the spreading of the pathogens to additional cells. Support for that notion can be found in the targeted killing of cells infected with viruses, whether in response to interferon or by T cells (although the latter is not "altruistic" per-se).

Based on these two examples of the direct link between inflammation and cell death, it is not trivial to provide a single explanation that covers both phenomena. One could even wonder whether these two processes actually share an evolutionary basis, or if it is merely an unusual example of convergent evolution. Evidence to link these axes, such as shared proteins, could shed light on their evolutionary source, as well as the rationale behind their common functions.

https://www.nature.com/articles/nri3561
https://academic.oup.com/carcin/article/30/7/1073/2477107
https://www.nature.com/articles/nature09782

We are all EU-Karyotes

When science and politics march together

By Ines Alvarez Rodrigo

Ines Alvarez Rodrigo is a DPhil student in Jordan Raff's research group at the Sir William Dunn School of Pathology.

Aristotle believed that men are 'political animals'. This has never felt truer in the UK than in the last few months, with Brexit *almost* happening twice by a margin of just a few days. The country as a whole has held their breath, incredulous as politicians argued, voted and got nowhere. As the timeline of events will be familiar to many readers, this article will not aim to give a detailed account of the UK's attempt to exit the EU. Instead, I'd rather explain the reasoning that led a 4th year DPhil student from Spain to stand in the centre of London for hours on Saturday 23rd March 2019 as part of the 'People's Vote March'.

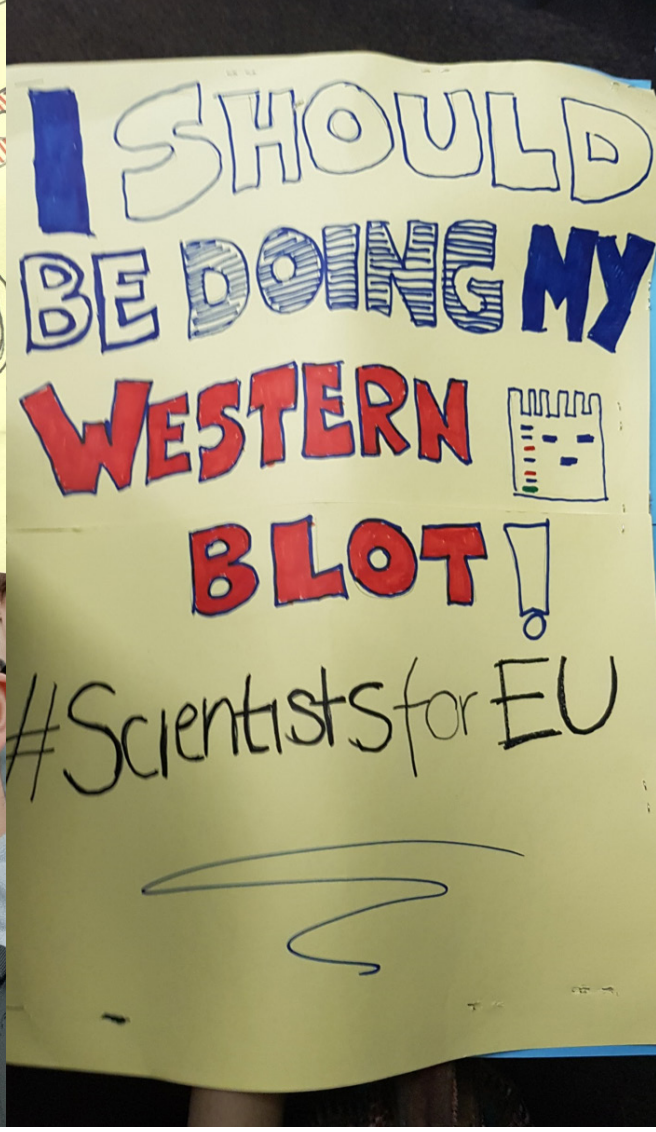
Before we continue any further, full disclaimer! This is an opinion piece. I am not a journalist, and I don't have the responsibility of giving a balanced view of both sides of the argument. Furthermore, I believe that sometimes an honest, truthful debate of an issue requires that not all sides are represented equally, because not all sides deserve the same platform.

I could write a whole article about the negative consequences of Brexit and why scientists should care about it. Briefly, Brexit will affect all aspects of our lives as individuals: from the availability of certain medicines (1) to the regular household income (2). However, because science is nowadays a global, collaborative enterprise, Brexit also presents specific challenges to the UK's scientific community. European scientists that enjoyed their rights to live and work freely in the UK might now feel unwelcome and unwanted. Many will consider moving elsewhere, just as some EU agencies (such as the European Medicines Agency) have already done. Research funding will also be lost, as the UK received more funding from the EU than it contributed (3). Finally, the Brexit campaign relied on emotional arguments ('take back control') and anti-expert sentiments. This philosophy is completely opposite to that of scientists, whose job it is to be experts in their field of study and to base their knowledge on the accumulated facts and evidence.

I am going to assume that this is a good enough (albeit short) summary of why a scientist might be against Brexit. The next question, though, is a bit trickier: why would anyone have marched on March 23rd? If 'Brexit means Brexit' and it is truly the 'Will of the People', why waste our time fighting a battle that cannot be won? The answer in brief is that, as has been proven ever since, the fight is far from over. In fact, I think there are three strong, *scientific* arguments why you should forfeit your Saturday plans next time that there is a pro-EU march:

1. Democracy and maths: We are being told repeatedly that Brexit is what the majority wants. But if you know basic maths, you know that this is not quite true. While Leave won by 51.9% to 48.1%, the referendum turnout was approximately 72%, and not everyone who lives in the UK was allowed to vote. This means that out of 66 million people living in the UK, only 17.4 million voted Leave. Yes, this is still more than the 16.1 million that voted to remain in the EU. However, 'Remain' is a single option, whereas (as has been proven by the chaos in Parliament) Brexit can in fact mean many very different things: a Norway model, a Canada-style agreement, no deal... A margin of 3.8% in the referendum and just 1.3 million people is too small to think that there are more people who support any single Brexit option compared to those who support Remain. Incidentally, approximately one million people support Remain so strongly that they were willing to not only vote in the 2016 referendum, but also march through London in 2019. If one million people are enough for our representatives to trigger the Brexit process, one million marching through London should give them pause. Additionally, if you (like me) were not allowed to vote in the referendum despite having made the UK your home, then marching is the only option to make your voice be counted.

2. The prisoner's dilemma: People who don't participate in demonstrations will often argue that it is not worth spending time and money attending them, because politicians are unlikely to listen unless they are a breakout success. The truth is that one can rarely be sure that a demonstration will be successful until you



arrive at the meeting point and see how many people are already there. Thus, attending a demonstration is a bit like the famous example of game theory, where prisoners "A" and "B" decide whether to stay silent or blame the other. If both prisoners cooperate and stay silent, the outcome will be better for them than if they blame each other. But the temptation to talk remains because if "A" blames "B" and "B" stays silent, "B" takes all the punishment and "A" goes free. Similarly, if everyone makes the effort to attend, the demonstration will make the headlines and show that their opinion is worth listening to (i.e. positive outcome via cooperation). It is also possible that you stay home and the demonstration succeeds nonetheless, in which case other people put in the effort but you reap all the benefits. However, if everyone stays home, the march will certainly fail and the message that politicians will receive is that not enough citizens care about this issue.

3. Power to the scientists: 30 years from today (climate change notwithstanding), if you are asked about what you did during the Brexit process, what will you reply? Some people will argue that attending a demonstration is always worth it. There is a chance that their efforts will pay off in the end, but even if this is not the case, at least they have done everything in their power to change society within the democratic system we live in. In this sense, demonstrations give scientists the opportunity to become a visible 'lobby' and make their opinions be heard as a group (e.g. Scientists for EU). We can also challenge the stereotypes that some sectors of society still hold against scientists, viewing them as old, crazy, white men that don't care about the lives of regular people or the world outside their lab.

Overall, attending the 'People's Vote March' on the 23rd March was an extremely positive experience. I felt welcome and valued as a member of society, and the atmosphere was very respectful and family-friendly. We marched in the company of fellow scientists, we chanted and we saw great placards (including one with the EU-karyote pun from the title, which I cannot claim as my own). I even had the opportunity to do a bit of public engagement, trying to explain to a fellow marcher what a western blot is. From the moment that my lab mate Ed and I got on the packed train bound to London, I knew that attending the march was the right decision. The events that followed that day have only strengthened this belief: we have gone from a hard Brexit bound to happen on the 29th March, to a 'flextension' and serious possibilities of a softer Brexit or no Brexit at all. Only time will tell, but what we can all be sure about is that there will be more pro-EU demonstrations to come, and I invite my fellow scientists to join me and others and bring their best scientific puns.

1. Pharmaceutical Services Negotiating Committee (2018) Response to the 'Informal consultation on urgent changes to the Human Medicines Regulations 2012 to ensure the continuity of supply of medicines (including in a 'no deal' Brexit)'. Available at https://psnc.org.uk/psncs-work/psnc-publications-and-resources/responses-to-consultations/ [Accessed 21st April 2018]
2. Smith J (2019) Counting the cost: UK living standards since the 2016 referendum. Available at: https://www.resolutionfoundation.org/publications/counting-the-cost/ [Accessed 21st April 2018]
3. Royal Society (2015) UK research and the European Union: The role of the EU in funding UK research. Available at: https://royalsociety.org/topics-policy/projects/uk-research-and-european-union/role-of-EU-in-funding-UK-research/uk-and-eu-research-funding/ [Accessed 21st April 2018]

What Can Dinosaurs and Darwin tell us about Modern Capitalism?

By Sunetra Senior

Sunetra is a freelance writer and editorial executive known for her creative flair. She is a notable features columnist for Asian Business Publications, and writes extensively on feminism, social democracy, and the psychological nuances of modern culture. She has a MA in English Literature from Warwick University.

Dinosaurs provide us with a perfect example of dominant Darwinian evolution: separated from humanity by approximately 230 million years as well as, possibly, a giant, errant meteorite. This allows us a distinct perspective on our personal evolution as a species, namely on the anthropological aspect of 'survival of the fittest', and the current model of capitalism upon which this is based. Here, the popular sentiment is eloquently summarised by biological researchers, George Constable and Tim Rogers, from the Universities of Zurich and Bath, respectively, in their piece, *The Selfish Myth:* "From rivalry in business to the adversarial focus of the mainstream media, competition appears to be king. In the cutthroat world of evolution by natural selection, if I can exploit somebody to improve my own lot (and have a chance to pass on my genes to future generations) then I should take it…In the end, the population should be genetically predisposed to exploit one another, undeterred by long term negative consequences."

Their paper states that the natural world actually demonstrates as many instances of species evolving to help, bettering the chances of their mutual survival: "co-operation and altruism are in fact quite common…from apes to honey bees. Amazingly, even some single celled organisms have evolved to be altruistic. Brewer's yeast (the stuff that makes your beer fizzy) can excrete enzymes that convert chemicals in the environment into food. This provides sustenance for itself and the surrounding cells so the colony can grow." As a corollary, the prehistoric paradigm of dinosaurs allows us to take an immense, conceptual stride further: not only understanding that the value of collaboration is underappreciated in the development of humanity, but, moreover, that the vernacular of ethical superiority commonly surrounding such conversation is, itself, flawed, reflecting a yet deeper, corrosive practice.

It is no coincidence that the largest dinosaur discovered to date, is a Titanosaur, formally dubbed *Patagotitan mayorum*, which belonged to a group of sociable, herbivorous sauropods. It was these gentle giants who were technically winning the 'Dino Arms Race' before a lump of icy rock, if we accept the extrinsic catastrophic theory, collided with Earth, resulting in the dinosaurs' extinction. It was not the infamous dagger-toothed Tyrannosaur who emerged virtually unassailable, but the placid Titanosaurs who steadily roamed the planet and preferred to look after their own. Their size and communal nature rendered them essentially beyond the fatal attacks of even the largest and most co-ordinated of carnivores.

It is worth noting, then, that it was the more nuanced habits of cooperation and care, which determined the organic strength of these Titanosaurs, in a world increasingly characterised by titans. It was necessary that sauropods grew larger as a defence against the aggressive Tyrannosaurs of the era, who could inflict potentially irreversible damage on their prey. Thus, a more profound biological truth emerges. Despite being seemingly at odds, there was, actu-

ally, an underlying connection between predator and prey, where the two were mutually dependent.

The same ecological equilibrium can be applied to humanity, which is conversely defined as a species through its intellect. Beginning with the most primitive Homo sapiens, who might have fashioned spears out of wood to defend themselves from animal attacks, humanity has advanced because of its mental acuity. As opposed to brute force and the many spectacular phenotypic adaptations of the dinosaurs, humanity's sophisticated infrastructure, dynamic communications, and synchronized bartering system, otherwise known as 'the economy', have allowed us to claim the majority of the planet via a more complex psychological interaction. However, here, examining the way our global society is developing, predominantly through the financial structure of globalisation, we see a disproportionate focus on the principles of competitiveness and the individual over the values of moderation and collective support. In his well-argued book, *Making Globalisation Work*, winner of the Nobel Prize for Economics, and former Chief Economist at the World Bank, Joseph Stiglitz, corroborates this world view. He writes on 'The pervasiveness of poverty,' 'The need for foreign assistance and debt relief', and 'The aspiration to make trade fair' as just a few of the factors, which demonstrate that the contemporary capitalism linking us, is in an unbalanced, destructive phase. However, it also has great promise to provide a universal boost: "trade liberalisation – opening up markets to the free flow of goods and services – was supposed to lead to growth... the most hotly contested policy issue of the 1990s was capital market liberalisation, opening up markets to the free flow of the short-term, hot, speculative money...By 2003, even the IMF (International Monetary Fund) had conceded that… for many developing countries, capital market liberalisation had led not to more growth, just to more instability."

This neoliberal volatility circles back to the developed countries themselves: "The IMF failed in its major mission of ensuring global financial stability – as evidenced so starkly in the global crisis at the end of the 1990s…globalisation has unleashed market forces that, by themselves, are so strong that governments, especially in the developing world, often cannot control them." So, why then, knowing this, do we still not fully recognise the need for stability? After all, we are witness to the intense environmental tumult, in the form of global warming and widespread social unrest. It is a reflection of misinformed, perhaps even *malformed*, power propaganda. A handful of corporate monopolies, which, presently, hold sway over the media and world politics refuses to systematically compromise: "Those who benefit from the current system will resist change, and they are very powerful."

The extreme control manifests itself through xenophobia, obsessive individualism, and an almost palpable 'Us-versus-Them' agenda that has, inevitably, spread throughout the international community, extensively distorting perception, and exploitatively dividing it. Here, we return to the simpler, yet significant, time of the dinosaurs. As their external environment catastrophically changed around them, the herbivores were among the first to die in mass numbers. This, in turn, meant the predators slowly starved to death, slumping along abandoned Titanosaur dust trails, searching for food. To pursue the analogy, humanity comes to be its own devastating comet. It is counter-productively depriving and killing off huge parts of the planet, from those seen to be lower in the hierarchy to the many species of plants and animals, on which the few at the top rely. The wider ecosystem will eventually buckle, quashing those responsible for the oppression too.

A constructive, timely, change in collective consciousness, or a smart mutation then, is the adaptive answer that rescues the human race. The peppered moth famously evolved overnight during the sooty challenge of the industrial revolution; today, humanity must mentally challenge a rapaciously illogical world. Indeed, to return to the visceral relationship between the two apex dinosaurs who coexisted, togetherness and antagonism must always work together as part of the greater natural order.

In truth, Darwin, a consummate scientist, never championed the idea 'of the survival of the fittest,' much less the concept of might as an unparalleled virtue. This perhaps, unsurprisingly, was actually the belief of an ultra-conservative sociologist, Herbert Spencer, who applied Darwin's theories to his economic predictions. Darwin objectively documented diversification and creative problem-solving as the signifiers of biological strength. Significantly then, Stiglitz argues in favour of fairer financial regulation, retaining the basic prototype of capitalism, but modifying it towards a more socially conscious design. Indeed, his meticulously composed global measures, both "small and big", could realistically curtail the rule of large multi-national corporates. This would allow intervention at crucial stages during economic hazard zones, more equitable business with developing nation-states, and a plethora of diverse businesses and skill-sets across the world to flourish. This enhances the livelihoods of everyone involved - overfed tyrants included - and truly pays homage to Darwin's original thesis.

In fact, to shun the empathetic knowledge that allows a clear evolutionary route to survival, not only undercuts the intelligence that defines us but, also, surely, perverts it: to the point where Darwin might even have classed it a terrestrial disease. Indeed, the unrelenting coldness and denial that comes of this are endemic. Hyper aggression agitates ideological conflict and creates blinding hypocrisies, needlessly turning different groups, sometimes violently, against each other. Thus, in recent times, compassion isn't simply a neglected aspect of nature, but a suppressed part of our genetic lineage, which must be consciously revived. As The Selfish Myth states: "Altruism tends to emerge in the long run." The true sign of success as defined by Darwin is not the fury of competition, but well-being for the many as the product of a fundamentally integrated world. The simple existence of vying polarities really only reflects the dazzling intricacy of such life.

1. Positive News, The Selfish Myth Issue 87, p.24 – 25
Co-authored by George Constable, Tim Rogers
2. Making Globalisation Work, Another World Is Possible, P.15-16, p.20, p.13 (in order of quotes listed, of which there should be 3). Published by Penguin, 2006. Joseph E. Stiglitz
3. http://www.bbc.co.uk/earth/story/20160201-meet-the-most-massive-dinosaur-to-ever-stomp-the-earth date visited: 25/04/19
4. https://www.smithsonianmag.com/smart-news/new-evidence-peppered-moths-changed-color-sync-industrial-revolution-180959282/ (Visited 27/04/19)
5. http://newlearningonline.com/new-learning/chapter-4/herbert-spencer-on-the-survival-of-the-fittest date (Visited 27/04/19)

SCIENCE COMMUNICATION IN THE FAKE NEWS ERA

Can we get 'good science' on the front page?

By Dr Anna Caballe

Dr Anna Caballe, Postdoctoral Research Associate at the Dunn School of Pathology and Wolfson College

Communicating science effectively and reaching wider audiences is always challenging for researchers. In the last few years, we have entered the age of fake news – with an information overload dominating the media channels and reliable science being the needle in the haystack. How is the public hearing about science? Can we get 'good science' on the front page?

A recent event in Oxford brought together a panel of experts with roles in academia, journalism, science communication/media, editing and policy, for an extremely interesting discussion about the challenges and realities of communicating science in the age of fake news (1). It is a topic that could be debated extensively, however, some key points raised by the experts are worth remarking.

The challenges when engaging the public with scientific research

Fake news influence the information received via different media channels and there are concerns of this phenomenon affecting the public science discourse. The umbrella of what is considered 'fake' regarding science encompasses a range of stories: those presented with bias or as propaganda, scientific papers given a disproportionate spin, unpublished data that somehow hit the headlines or, quite frequently, misinterpretation of scientific facts (often by non-specialised journalists). Remarkably, it appears that the amount of actually fake science news stories that are deliberately created is quite low.

Some argue that these types of stories are not new, that they have been around for many years. What is shifting,

"One of the biggest values of researchers engaging the public is their ability to communicate the crucial scientific method and why they are engaging in research"

however, is what kind of information is trusted and who are the 'truth-tellers' in the eyes of the public. Unfortunately, cases of sloppy science have led to the mistrust of experts or, as some would call them, 'the elite'. A study by Queen Mary University has recently found a link between the rise of populism and increasing vaccine scepticism (2). Science communication experts also agree that there are some lost battles when talking about, for example, vaccines or genetically modified organisms, whereby critical sectors of the public are unlikely to change their views – miscommunicated research and misleading conclusions based on scientific data are hard to challenge even with the best scientific evidence.

Unfortunately, cases of sloppy science have led to the mistrust of experts or, as some would call them, 'the elite'.

What is also rapidly changing is how information is shared and the platforms used to access this information. Despite a proliferation in scientific knowledge, with two million new articles published each year, only one-sentence headlines often make it through social media or fast online news. As people's attention-span is much shorter, getting complex messages and relevant stories across and above the noise of all the other news becomes harder! Nevertheless, it is encouraging to read that 83% of British respondents to the YouGov-Cambridge Globalism Project survey (a wide-ranging poll of >25,000 people on populism, globalism and technology) had little or no trust in the information of platforms like Facebook and Twitter (3). The real question is, how are the unreliable or poorly explained science news stories affecting behaviour? Public Health England carries out annual surveys to assess parental attitudes to child immunisation. Some of their recent figures are quite reassuring; 93% of parents agreed that health professionals are the most trusted source of advice on immunisation (4-5). However, work needs to continue to address the 4% who refused to vaccinate their children, a figure that might not be sufficient for achieving herd immunity; the resistance to the spread of a contagious disease within a population as a result of a sufficiently high proportion of individuals immune to the disease, especially through vaccination. Unfortunately, herd immunity is under thread in some European countries, where public attitude to vaccination has been shifting for the worse.

Despite a proliferation in scientific knowledge, with two million new articles published each year, only one-sentence headlines often make it through social media or fast online news.

Why should researchers actively engage in science communication?

It is well known that the public believes in researchers more than journalists and professional communicators, so it is important that they play an active role in science communication. One of the biggest values of researchers engaging the public is their ability to communicate the crucial scientific method and why they are engaging in research – nobody will be more passionate about this than the scientists performing the work. Scientists should also actively engage with the media and challenge inaccurate reporting, as author Dr Simon Singh has been doing with his Good Thinking charity, tackling bad practices in the media, healthcare and education systems, and dishonest health campaigns. Publishers, funding bodies and institutions should also provide the tools for researchers to learn how to communicate science and convey complex messages. They should also allow them the space to do this – the pressures of publishing and getting grants sometimes get in the way. In addition, the way scientific studies are made available to the public should be discussed. Although open access is gaining territory, there are still many papers behind paywalls and, even if they were all to be removed, most people lack the knowledge to critically analyse a scientific paper.

Perhaps we should go back to the basics and improve the education on scientific methodology and data interpretation at school-level. For the time being, let's nurture specialised science journalists and use everyday communication (from social media channels to an over-the-fence chat with the neighbour or the hairdresser) to talk about science with as many individuals as possible. And no need to wait until the paper is out, sharing the working hypothesis and method is part of the journey, as long as it is clearly explained that it is work in progress!

Publishers, funding bodies and institutions should also provide the tools for researchers to learn how to communicate science and convey complex messages.

1. Robert A *et al.* (2019) Communicating science in the age of fake news. Grand Challenges Seminar Series, Christchurch College, NERC DTP, University of Oxford.
2. Kennedy J (2019) Populist politics and vaccine hesitancy in Western Europe: an analysis of national-level data. Eur J Public Health, ckz004.
3. Hern A (2019) The new populism: Britons less trusting of social media than other major nations. Available at www.theguardian.com/world/2019/may/03/britons-less-trusting-of-social-media-than-other-major-nations-facebook-twitter [Accessed 14th May 2019]
4. Public Health England press office (2019) PHE offers support to UK vaccine heroes. Available at https://www.gov.uk/government/news/phe-offers-support-to-uk-vaccine-heroes [Accessed 14th May 2019]
5. Official Statistics. Quarterly vaccination coverage statistics for children aged up to five years in the UK (COVER programme). Health Protection Report, V.13 Number 11. Available at www.gov.uk/government/statistics/cover-of-vaccination-evaluated-rapidly-cover-programme-2018-to-2019-quarterly-data [Accessed 14th May 2019]

THE FUTURE OF LAB AUTOMATION

By Heather Jeffery

Heather Jeffery is a DPhil student in Conrad Niduszynski's research group at the Sir William Dunn School of Pathology.

Technology and science have been developing alongside one another for many years (1), with lab automation a prime example of these two fields converging. The basic premise of lab automation – using machines to perform repetitive tasks in order to save time – still applies today, despite technological advancements and dramatic changes in the appearance and function of such machines. Automation has been successfully utilised across all scientific disciplines, although is currently most predominant within the life sciences field.

The increased use of lab automation may be partly due to greater awareness of the importance of reproducibility in scientific research (2). In addition to saving time, lab 'robots' can perform specific actions more accurately and reproducibly compared with standard manual lab work (3). Furthermore, lab automation can provide scientists with the capability to perform more complex, challenging experiments and also speed up research by allowing more time to focus on experimental design and data analysis. However, automation can be expensive, limited in terms of functional diversity and challenging to use.

Singer Instruments, a robotics company operating in the South-West of England since 1934, has developed a range of high-throughput robotics for use in the field of microbiology. A key feature of these robots is that they are designed to be highly functional, but still easy to use.

Within microbiology, which originated with the identification of micro-organisms in the late 17th century, both the practical applications and methodologies have been constantly evolving, in line with technical advancements. Microbes are now some of the most commonly-used model organisms, with E. coli and S. cerevisiae popular 'workhorses' for routine lab protocols, such as molecular cloning. However, the majority of practical cloning work is still performed manually, which is both laborious and inefficient.

The collection of lab robots produced by Singer Instruments allow the user to perform a variety of molecular cloning steps easily and efficiently; this includes picking individual colonies, moving colonies between plates, imaging, counting and analysing plates, and even performing tetrad dissections to

separate yeast spores. Their newest robot, PIXL, enables colonies to be selected based on a variety of characteristics, such as size, circularity, brightness and colour. Subsequently, specific colonies can be picked and plated on to multiple target plates. The PIXL robot offers a vast amount of possibilities for customisation, thus supporting a wide range of experimental set-ups.

A standard approach to replicating plates of yeast (or bacteria) involves manually transferring the micro-organism between plates using a piece of velvet. This is a low-throughput technique and can lead to experimental inaccuracies. Singer Instruments have developed a high-throughput robotic machine, called ROTOR, which facilitates easy replication of array plates, in either 96, 384, 1536 or 6144 density formats. This both saves the user a substantial amount of time and ensures accurate plating.

Imaging is an important step of a cloning protocol, however, it often only provides qualitative data. The Singer Instruments' PhenoBooth machine enables high-resolution imaging of colonies on a plate. Importantly, it also provides extensive quantitative data, generating plate summaries or colony-specific information using a variety of parameters, similar to the PIXL machine. In order to gather this data, the software for both PIXL and PhenoBooth performs colony detection and counting. The PhenoBooth software is also supported by a web application that provides additional data analysis functionality, such as replicate patterns and alternative experimental design set-ups.

Finally, Singer Instruments also make two tetrad dissection microscopes; the manual SporePlay microscope and the automated MSM400 microscope. By facilitating tetrad dissections – traditionally a complex and specialist microbiological technique – these microscopes make tetrad analysis and isolation of yeast strains a more accessible and user-friendly protocol.

These robots each have their own powerful applications and can also be used alongside one another to pick, screen and image microbes in a high-throughput manner. Given that high-throughput microbiology experiments are becoming increasingly common, particularly for genetic and chemical screens, machines like these will be critical for enabling these screens to be performed at a faster rate, with greater reliability and reproducibility.

1. Olsen K (2012) The First 110 years of laboratory automation: Technologies, applications, and the creative scientist. Journal of Laboratory Automation 17(6):469–480.
2. Rethlefson M, *et al.* (2018) Building capacity to encourage research reproducibility and #MakeResearchTrue. Journal of the Medical Library Association 106(1): 113–119.
3. Jessop-Fabre M & Sonnenschein N (2019) Improving reproducibility in synthetic biology. Frontiers in Bioengineering and Biotechnology 7.

PIXL Microbiology Colony Picker: Singer Instrument's newest robot, PIXL, performs colony picking of microbiological cultures with the red robotic arm and specialised software.

In conversation with the Vice Chancellor of Oxford

LOUISE MARY RICHARDSON

Louise Mary Richardson was interviewed by Sonia Muliyil (Phenotype Editor-in-chief), and Marina Kolesnichenko (Co-founder of Phenotype).

Louise Mary Richardson, the 295th Vice Chancellor of Oxford, is the first woman to hold this position in 900 years of the University's history. Louise Richardson is a world leading expert on terrorism and authored several highly acclaimed books, including "what terrorists want". This book has been hailed both by experts and book reviews as "groundbreaking", "brilliant", "myth-buster" and "the book everyone has been waiting for".

Before coming to Oxford in 2016, Louise Richardson served as Principal and Vice-Chancellor at the University of St. Andrews. Between 1989 and 2001, Professor Louise Richardson taught at Harvard College and Harvard Law school and in addition served on numerous committees concerned with undergraduate education, human rights and women's issues.

Louise Richardson holds numerous awards and prizes, both for her research and excellence in teaching. In addition, she received several honorary doctorates from top international Universities. She is a member of the American Academy of Arts and Sciences, the American Philosophical Society, the Academy of Social Sciences in the United Kingdom, an Honorary Member of the Royal Irish Academy and a Fellow of the Royal Society of Edinburgh.

Vice-Chancellor thank you so much for taking the time to meet with us. Today's topic is "Aspire to Inspire" and we would be grateful if you could tell us about your journey and if there was someone who inspired you.

Even though she did not know it, a woman who really inspired me was Mary Robinson. Mary Robinson subsequently became the first female President of Ireland. She was a fabulous President and remains a wonderful woman. She then went on to work at the UN and now runs a charity on climate change.

But it was much earlier that she inspired me. I was a secondary school student and I did a lot of babysitting, and so I ended up watching a whole lot of television. I was always very interested in politics, so I used to watch all the political shows. Mary Robinson was always the only woman on any of these shows. There was one called "Seven Days", and she was always the only woman who was ever party to the conversations, but she was always so much smarter, so much more articulate, and so much more thoughtful than anybody else in these discussions. I used to watch her and think that it is so unfair

> ***...we as institutions and as society need to make it much easier. We are not structured in a way at all to accommodate combining families and careers.***

- here is this amazing woman, and yet she is a minor figure in a marginalised party. I subsequently went to University where she was a Professor. She was again on television then on liberal issues like contraceptives. Contraception was illegal in Ireland when I was there as a university student, and she lobbied for its legalisation. I remember thinking - this woman is so able, and so smart, and here she is having this ridiculous argument. I just don't want to spend my life having arguments about things, which to me are so self-evident - that women should have a right to contraception. I want to have arguments about really important issues. So that really prodded me to leave Ireland.

What kept you motivated?

I went to school in rural Ireland. We were taught to iron men's shirts. We started term ironing mens' handkerchiefs, and at the end of the term we got to ironing their shirts. And woe betide who had any wrinkle in the collar! And I remember saying then that I am never marrying any man who expects me to iron his shirts! I had three brothers and three sisters. There is nothing like having three brothers to explode the myth of male superiority. At school the aspiration was you married and became a housewife… I remember again, babysitting and watching a politician I admired, Austin Currie, on a program called the "Late Late Show". He said he studied History and Politics at university and I said that's it, that's what I am going to do! I am going to study History and Politics. I applied to Trinity College… Trinity was where Protestants went and I was Catholic. I was fairly rebellious and going to university and going to Trinity was part of my rebellion.

How did you transition from a solely academic career to VC?

The hardest thing I ever did was being an academic and having three young children at the same time. You learn to be very very good at managing your time and being really efficient. To this day, I am extremely efficient with my time.

When I was at Harvard my colleagues would make fun of me because when I would heat my coffee in the microwave I would always push 222. They would say - you are so fussy about the precise heat! And I said no - it is just much faster to push 2 three times.

You become very experienced with juggling a lot. I was gradually given more and more responsibilities running things, and I found that I actually quite enjoyed that. You got immediate results, rather than spending weeks and weeks pursuing something in the archive and maybe finding it or maybe not. Here I could actually find a problem and solve it. My career was not planned. I never imagined living in England and being Vice Chancellor.

Do you think mentors are important?

Everybody says they are, but I did not have a mentor. I absolutely, adored my thesis supervisor - Stanley Hoffmann. And certainly he wrote good references for me when I was looking for jobs but he did not mentor me in any way. I remember the time I gave him the first 150 pages of my dissertation. I delivered it to his office and I was a nervous wreck. Later, he said "I've read your chapters so you can collect them from my office." I raced to his office, I got the chapters …and I went through the whole thing and he had corrected two typos. I eventually found him and said - did you get the chapters - what did you think of them, you did not say anything. He said - I don't believe in giving faint praise. No he was not a mentor in a classical sense - he was a man who inspired me because he had a fabulous intellect and such an appreciation of history, culture, of the arts. But I did not have somebody advising me. I could have used that, I suppose. You make mistakes because you don't have advisors.

Do you think it makes you stronger? More independent?

I would suppose so. I would still advocate giving people mentors. The thing that troubles me most is just how hard it is to combine [career] and a family. I don't mean to talk about children all the time, as if they were only a woman's issue. But predominantly they are. I look back on that time and I have no idea how I did it. Writing lectures for the first time as a junior academic is so much work. Subsequent years you can just update the lectures but first time around it is brutal. I filed my dissertation on the day my daughter was due. So I had no break, and then I started working immediately. So it was all very difficult. The pressure of trying to stay up all night writing those lectures to try to get them done before she woke up in the morning. Things afterwards had been so much easier. But the thing is - we lose so many wonderful women at these early stages of their career for completely understandable reasons. I totally understand people making other decisions. But I think, we as institutions and as society need to make it much easier. We are not structured in a way at all to accommodate combining families and careers.

Do you have any advice for any young career for women/men?

What I say to people is try to think of your career in the long term. When I was a junior faculty member at Harvard, which is an unbelievably competitive place … my goal was to keep my career alive while I had my kids. And I worked unbelievably hard. I just had different standards for myself at different times. My goal was just to get through these years keeping my career alive, so that when the kids were at school and I had more time, my career could take off again. Many people see themselves falling behind because they are distracted with other things and feel they are not going to make it. But they are thinking in the short term - you've got to think of your whole career.

THE JOURNEY OF A FIRST-GENERATION GRADUATE

By Dr Martine Abboud

Dr Martine Abboud is a Junior Research Fellow at Kellogg College and works at the Department of Chemistry.

I am driven by curiosity.

I grew up asking my parents loads of questions about everything around us. I was so fascinated by the stars and galaxies that I wanted to become an astronaut. However, during my teenage years my grandfather was diagnosed with cancer. He was one of the closest people to my heart, and his illness made me question my career choices. I wanted to help people but did not feel suited to working in a hospital, so I decided to pursue a career in scientific research. At the undergraduate level, I started by learning biology to better understand physiological processes and their pathological implications. Soon after, I realised that Biology and Chemistry are complementary and that an understanding of both fields is important to achieve results of clinical relevance. Hence, I opted for a secondary focus in Chemistry, both at the Lebanese American University. The doctoral programme in Chemical Biology at the University of Oxford caught my attention with its interdisciplinary nature. Coming from a minority background, I feared applying to Oxford because of how competitive and prestigious it is, but my mother was right – not applying is a definite rejection. I am glad I did. Five years past this day, I am featured on the Forbes 30 Under 30 list for my work to date.

Currently, I am a Junior Research Fellow at Kellogg College, Oxford. During my time at Oxford, I worked at the interface of Biology, Chemistry, and Biophysics. I think basic research is important in understanding molecular mechanisms and I have enjoyed doing both proof-of-principle and applied studies. My research has led to novel method development and advances in three different areas of scientific research. These include oxygen sensing, antibiotic resistance, and metabolic alterations involved in cancer. My research has been internationally recognised from both academia and industry. I am interested in enabling science, community, and policy to combat scientific misconceptions. I aspire to meaningfully contribute to society.

For details about my work, see this spotlight in The Oxford Scientist (http://oxsci.org/2019/02/19/spotlight-dr-martine-abboud/).

"Coming from a minority background, I feared applying to Oxford because of how competitive and prestigious it is, but my mother was right – not applying is a definite rejection."

Today, I would like to share the lessons that I have learned during this journey. These enabled me to grow both as a scientist and as a human being.

• Not applying is a definite rejection – do not be afraid to try.

• Academic merit is as equally important as being kind. No matter how good of a scientist you are, always treat other people well. This is the decent thing to do.

• Biology and Chemistry are complementary. We are in the era of interdisciplinary and collaborative science. Talk to scientists in other fields; they will have new perspectives. Always remember that collaborations, rather than competitions, drive science forward more efficiently.

• Being great at science and at people and/or money management are not necessarily related. Do not shy away from getting the trainings you might need as a scientist. All these skills and trainings will pay off.

• It is important to troubleshoot all the time. Some of the most exciting discoveries in science arise from mistakes. Research is full of failures on a daily basis; enjoy the small successes while understanding what went wrong, and why it did.

• Success in science is not an overnight effort, it is the accumulation of years of hard work. Be persistent.

• Science has no nationality, it is important to build bridges with underdeveloped countries.

• Science and art are complementary on many levels. Science is nature being artistic.

• A scientist is a human being who has hobbies too; we therefore need to humanise scientists to the public.

• Scientists are not meant to be lifetime technicians. Learning how to be creative in a technological era is crucial.

• As young women we are more prone to implicit bias. Do not be afraid to be assertive and reach out to more experienced peers for help and network building.

• The current culture of 'post-doctoral nomading' is destabilising for people with caring responsibilities. It should not feel like an obligation. We need better environments that allow us to thrive.

• Proper life-work balance is important and nurturing; it enhances productivity and happiness.

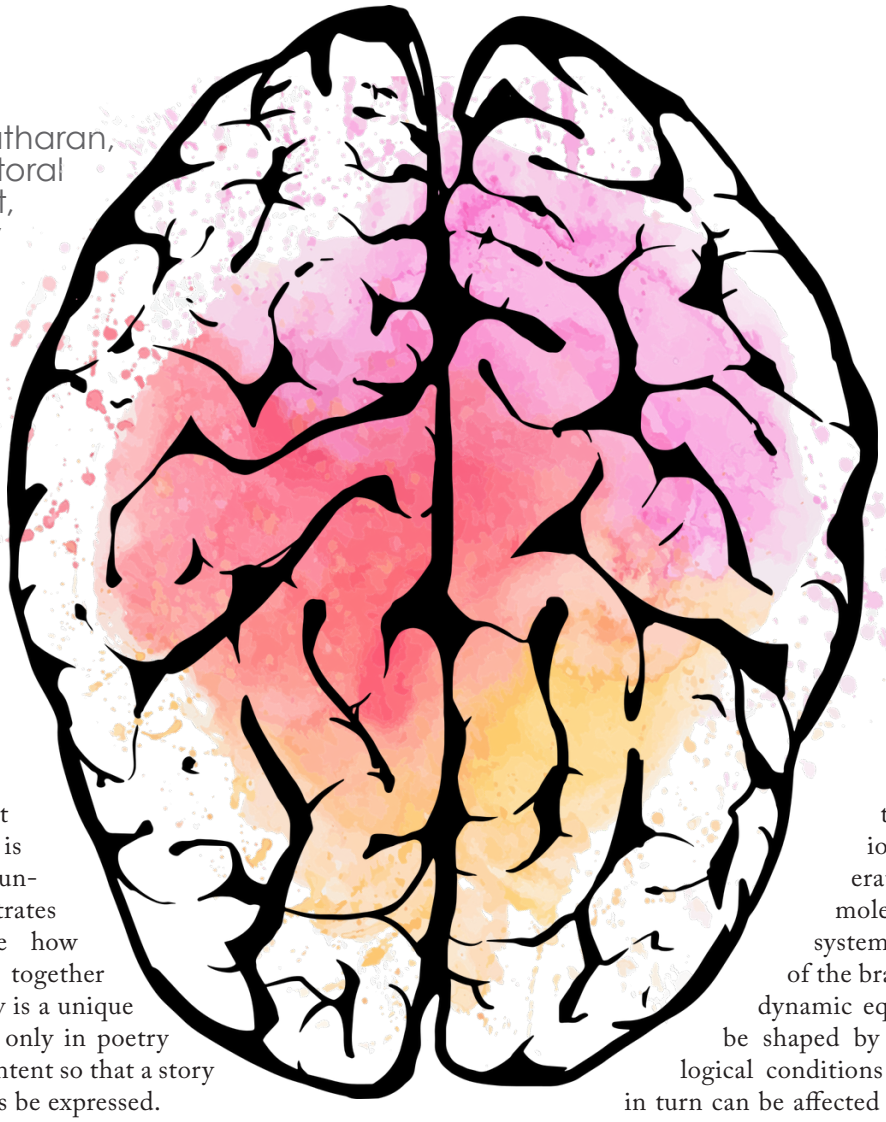
neuroscience & poetry

Discovering the neuropoet within you!

What is poetry? Is it an idea? Or is it a feeling?
Does it have a definition? Or does it defy definition?

By Jayanthiny Kangatharan

Jayanthiny Kangatharan, PhD, is a Postdoctoral Research Assistant, Harvard University



'Poetry cannot be defined, only experienced' Christopher Logue once remarked. Along the same lines, William Wordsworth suggested that 'poetry is the spontaneous overflow of powerful feelings'. There may be no clear-cut definition of what poetry is but what is clear is that unlike prose, poetry illustrates visually using structure how form and content come together to create meaning. Poetry is a unique literary art form as it is only in poetry that form does act out content so that a story can be told, and emotions be expressed.

What is most remarkable to someone like me, who studies the mind and the brain, is that poetry is a fascinating way to express human experience and understand human nature. Among all sciences, it is neuroscience that aims to understand how our nervous system operates, and how nerve impulses transmit information that allow our brain to perceive, think and learn. The human brain has many levels of organisation, spanning molecules, synapses, neurons, circuits, networks, layers, maps and systems, which ultimately give rise to our thoughts and perception. By the same token, thoughts and behaviour can affect operations at the genetic, molecular, cellular, and systems level. Thus, all parts of the brain work together in a dynamic equilibrium, which can be shaped by an array of psychological conditions and behaviours that in turn can be affected by environmental influences. In this regard, neuroscience is special, as it presents an amalgamation of different sciences that allow the brain to study itself.

What do poetry and neuroscience have in common? Both poetry and neuroscience throw light on many shared aspects that we can explore to broaden our horizon of human nature and to make sense of our human existence. One such aspect

"Poetry is a widening of consciousness, an extension of humanity."

is consciousness. Neuroscience is the field that attempts to find answers to questions such as how our brain produces conscious thought. Neuroscience research therefore explores this connection between our conscious experience of life and our human nervous system, thereby trying to understand what happens if our normal conscious experience is somehow diminished or disrupted. According to the poet David Constantine 'poetry is a widening of consciousness, an extension of humanity.' When we view life, in the words of Remy de Gourmont, as a 'series of sensations, each connected to a different state of consciousness', then it becomes clear that both poetry and neuroscience try to help us to understand the meaning of our human existence.

As a scientist and someone who grew up with a love for music and the visual arts, I have always been interested in ideas that would allow me to bring art and science together as it is only through creative and innovative endeavours that scientists are given the opportunity to express their artistic voice and that artists are given the opportunity to express their scientific voice. I was keen on understanding human nature using poetry as a tool while looking at discernible instances of human experience through a neuroscientific lens. This attempt to fuse poetry with neuroscience manifested itself in the idea of neuropoetry. Neither the term nor the idea are new as in the most general sense we all have been engaging in the act of expressing ourselves poetically in our desire to understand ourselves better. In the most specific sense, neuropoetry can be defined as poetry of the mind and the brain, which deals with a particular neuroscientific phenomenon such as a neurological condition or, which tries to convey specific processes at the molecular, cellular, synaptic, or neuronal level.

This attempt of neuropoetry to combine art and science inspired me to create neuropoems and take neuropoetry up a notch: I started to construct neuropoems as a riddle the answer to which reveals the subject of the poem itself. By adding this extra layer to the intellectual complexity of a neuropoem, I tried to encourage the reader to actively think about the content and enter an interactive dialogue with the neuropoem to arrive at the answer. With neuropoetry I was therefore keen on empowering the reader to learn more about neuroscience and delve deeper into the study of a certain neuroscientific topic.

To this end I launched a collection of such neuropoems during Brain Awareness Week in March known as 'Multilingual Neuropoetry'. I created the neuropoems in ten different languages to reach a wider audience. It is said that a poem lives in its language, which is body to its soul. According to the poet Joseph Brodsky, 'poetry is essentially the soul's search for its release in language'. By expressing neuropoetry in more than one language I wanted to create different bodies through which the soul can express its unique facets differently. The neuropoems in the collection are written in the languages Tamil, German, English, Latin, Spanish, French, Ancient Greek, Mandarin Chinese, Arabic and Italian. Translations are provided after each non-English poem, and each poem is accompanied by notes that elucidate the connection between the specific neuroscientific topic and the particular poetry form, in which it is presented. By using multiple languages, I also hoped to encourage everyone to take up a new language. Not only will you expand your cultural horizon or advance your career by doing so, but you will equip yourself with a larger cognitive reserve (1). While creating the neuropoems, my love for nature inspired me to come with a new poem form that I call the rainbow poem, which, similar to the rainbow that has seven colours, is seven lines long. Each line starts with the first letter of each of the seven rainbow colours.

With 'Multilingual Neuropoetry' I aim to show that poetry and neuroscience ultimately try to help us to understand human nature, and that science and art are essentially not that different in this regard. There is indeed evidence of the direct effect that poetry has on our nervous system, showing that engaging in poetry stimulates our emotional development and intellectual growth as we are using language to symbolically represent our experience (2). Writing poetry can also have many health benefits: by helping us to articulate our emotions and heal through self-expression, writing poetry can help us to understand our own feelings (3, 4). Apart from these therapeutic effects, writing poetry yields the obvious cognitive benefits because it helps you to understand language better, and challenges your critical and creative thinking (5).

I believe there is a poet in each and every one of us, and with this collection I would like to encourage you to use poetry as a way to express how your brain makes you feel. To support such endeavours that help art and science to join hands, and for information on the book and neuropoetry events, please check out the blog themultilingualbrain.blogspot.com, and like the Neuropoetry facebook page. If you are interested in organising a neuropoetry workshop in your area, please do not hesitate to get in touch with me at: jayanthinykangatharan@gmail.com.

SNAPSHOT

Congrats to Robert Lees for winning our Hilary 2019 Snapshot cover contest!

The cover photo was created from images of the whisker-responsive area of somatosensory cortex taken through a cranial window in a living mouse. This mouse in particular is transgenically-expressing GCaMP6s, a calcium indicator (false-coloured green), in almost all its neurons in the cortex.

There is a single high resolution image in the background of a single layer of neurons that was stitched together from multiple smaller images taken using two-photon imaging. The other colours overlaid on top of this background image correspond to average responses of the calcium indicator to stimulation of different individual whiskers, each whisker corresponding to a different colour.

Overall, this image shows the crude mapping of individual whiskers to somatosensory cortex in two areas (S1 and S2). The areas are mirrored and so each colour is present on both sides of the window, S1 is on the right, S2 is on the left. We assess this topography for later experiments where we want to induce activity in downstream areas (e.g. S2) from stimulation of a defined group of individual cells in the upstream area (e.g. S1).

The image looks as though it was taken in the darkness on an exotic planet, looking up at the cosmos through tree branches.

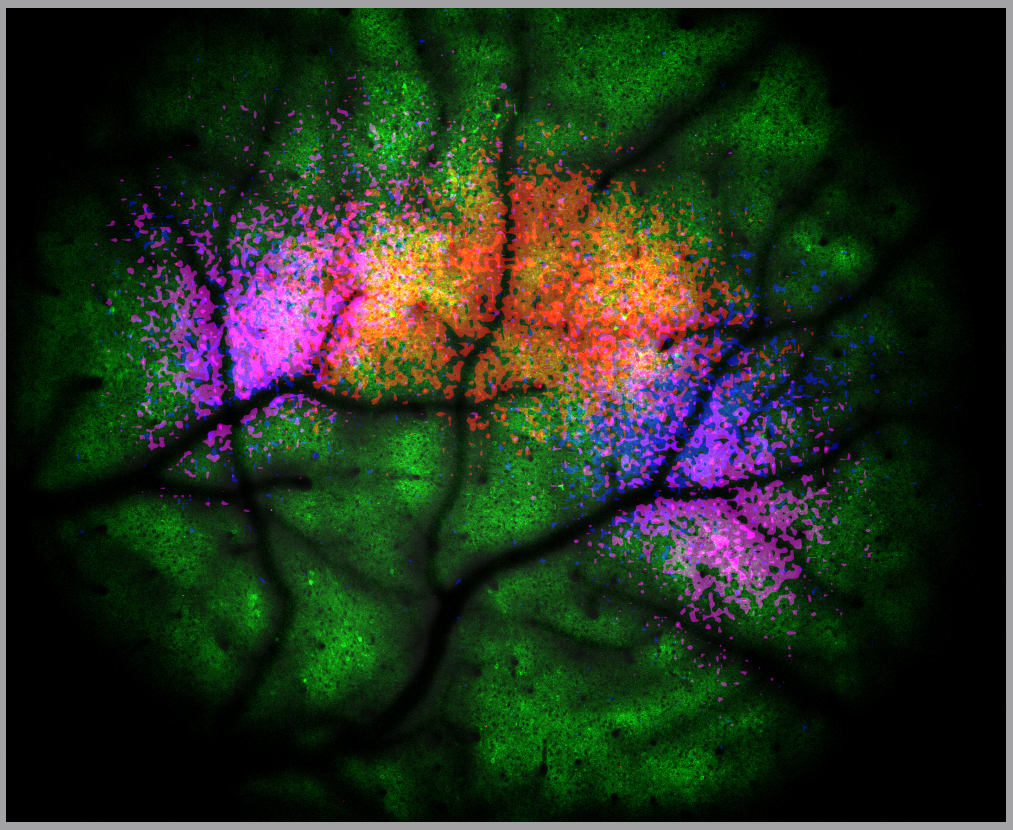
You could be our next Snapshot winner!

Send us your high quality scientific photos for a chance to be featured on the next cover of Phenotype

Pitch us your illustrations

We are looking for original artwork to accompany our articles

Entries should go to oxphenotype@googlemail.com



"The image looks as though it was taken in the darkness on an exotic planet, looking up at the cosmos through tree branches."

I am interested in mechanisms within the brain that allow us to perceive the world around us. The transfer of neuronal activity from one cortical area to a connected downstream area is thought to be an essential mechanism for perception. I use mice as a model to investigate the ways neuronal activity from one set of neurons can be transformed to elicit activity in another set of neurons in a downstream area. The first step to achieve this is to locate the areas of interest, in mice this involves stimulating a single whisker and finding the neuronal response in the primary area and connected downstream area. For example, the cover image is the colour-coded response for three separate whiskers, each showing two defined patches of response (primary and downstream) in the brain. I then define groups of individual neurons in the primary area to stimulate in physiologically-relevant or random configurations (both spatial and temporal) to attempt to find an efficient way of eliciting neuronal activity in the downstream area. This work will enable a more thorough understanding of how the cortex functions to encode perception, leading to more informed computational models of the brain that may inspire future artificial intelligence or brain-machine interfaces.

Dear Reader,

Writing this short piece, I feel as excited and nervous as I did while composing the address in the very first issue of Phenotype magazine. Phenotype was initially intended to be a newsletter produced by the Oxford University Biochemical Society (OUBS) for the students and post-docs in the department. I was President of the OUBS from 2008 to 2009 and, together with the core committee, a founder of Phenotype. Apart from Nick Anthis, a well-known blogger, none of us had much experience in writing or editing at the time. Sarah Iqbal and David Yadin, who took over editorial responsibilities, and greatly contributed to success of the journal, later joined us.

Looking at current issues of Phenotype, it is great to see that some of the original features remain, including the '5' with' interview style article, the crossword puzzle and the address from the OUBS President (nowadays, the Editor-in-Chief). But the journal has also evolved and flourished over time and has even been extended to other life sciences departments within Oxford!

Now, a decade later, I want to share 10 fun facts about Phenotype with our readers, so you can better understand the origins of the journal:

1. The name 'Phenotype' is a play on words and was conceived by the Secretary of OUBS, Maria Demidova. The initial designation of the journal was simply 'OUBS Newsletter'. However, Maria struck gold during a brainstorming session when she came up with the name 'Phenotype'; we were going for something that would not only capture the writing aspect, but also combine it with a scientific element.

2. First issues of the journal were written and put together by the OUBS committee. The first issues of the journal are signed by the entire committee to reflect the teamwork that went into producing Phenotype: Maria put together the crossword puzzles; Nick Anthis and Alice Blachford contributed original articles; and I wrote the introduction, conducted interviews and contributed photos. Everyone on the committee played an important role in the establishment and production of the journal.

3. The 1st committee had seven women. By 2009, our committee had grown to a total of seven officers: Maria Demidova, Muhan Wang, Pellin Ulluocak, Camilla Oxley, Maria Carrol, Sarah Iqbal and myself. In addition, two 'post-doc representatives' (Nick Anthis and Rodrigo Reyes) and an 'undergraduate representative' (Alice Blachford) became part of our team.

4. The journal was one of three main projects that we carried out that year. The first was a lecture given by the Nobel Laureate, Andrew Fire that was attended by over 300 people. The other project was the annual Biochemistry Black Tie Dinner, which was organized by Maria Carroll (the Social Secretary of OUBS).

5. The '5' with' article was initially meant to be a regular interview piece with the Director of Graduate Studies, Mary Gregoriou. Although Mary was not officially an OUBS member, she was an invaluable source of support; if you were not sure whether you could get away with American spelling in your thesis, if you needed to check whether you should wear full sub-fusc for a transfer report viva, or if you simply needed emotional support, Mary G was always there for graduate students. Therefore we wanted to set up '5' with' in an interview format so that 'a lost first year DPhil' could ask Mary questions about the procedures and regulations in the department. That title, however, went to a different feature and the interview with Mary was renamed to 'Graduate Matters'.

6. '5' with' was also a play on words. Some of us were working on 3' and 5' regions of RNA and got carried away!

7. Issue 1 of Phenotype was primarily available online. As we had very limited funds and getting sponsors in the beginning proved difficult, Phenotype was initially only set up online. However, a couple of our committee members had access to colour printers and managed to print out A4 copies to leave around the department.

8. To increase our readership, we offered prizes to those solving the crossword puzzle in the first issue. Initially we thought of offering free pints of beer at the University Club, but in the end we offered a book voucher for £10 instead!

9. The original mission of Phenotype was to inform the department about the events organized by the OUBS and to encourage scientific discussion. Part of the reason for creation of the journal was to give us, as a society, more visibility and attract new sponsors. Phenotype is now independent of the OUBS, nonetheless the mission to encourage scientific discussion still holds true.

10. After leaving OUBS and Phenotype, we are still in touch. Half of us ended up moving to the USA for post-doctoral work. I am happy to say that I am still in touch with some of the Phenotype alumni and working together as a team was one of the best experiences I had in Oxford.

Sincerely,

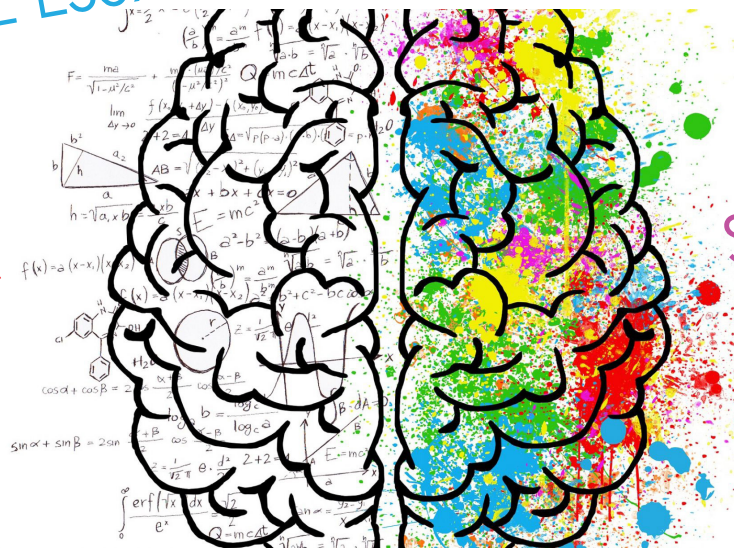
Marina Kolesnichenko, a co-founder of Phenotype

Embrace your creative side

Phenotype wants to publish your *science* themed pieces of creative writing.

PERSONAL ESSAYS

SCREENWRITING



POEMS

SHORT STORIES

Send pitches and submissions to oxphenotype@googlemail.com

Past founders of *Where are they*

The OUBS committee with Nobel Laureate Andrew Fire.

Muhan Wang (Treasurer 2008-9): I am currently working in Unilever R&D, as product development manager for personal care. One of my fondest memory was organising the Nobel Laureate Lectures.

Marina Kolesnichenko (President 2008-09): After completing the joint Oxford/TSRI DPhil/PhD program, I moved from the USA to Germany. I am currently a postdoctoral researcher at the Max Delbrueck Center for Molecular Medicine in Berlin. My fondest memory of Phenotype was the brainstorming sessions. At one point we were trying to outdo each other and come up with the most ridiculous titles possible – "Mary G's Jewels of Wisdom" was a suggestion for what later was called Graduate matters.

PHENOTYPE *now?*

Camilla Oxley (IT Officer 2008-09): After 4 fun years in Oxford getting my PhD in Biochemistry and participating in the foundation of Phenotype, I moved to the US for a 4 year postdoc in cancer research at the University in Pennsylvania. Now I work in drug development at Johnson & Johnson where I take molecules from discovery to clinical trials and love going to work every day.

Nick Anthis (Post-doc Representative 2008-09): After Oxford, I was in the Washington, DC, area as a postdoc at NIH and then a science policy fellow at USAID, before moving to Oakland, CA, in 2016 to work for the University of California Office of the President (UCOP). I'm currently a program officer for the California Breast Cancer Research Program, a research funding program based at UCOP. My work there involves developing funding opportunities, managing the scientific review of grant applications, and overseeing a portfolio of research awards. I served as treasurer of the Oxford University Biochemical Society (OUBS) from 2006 to 2008. During that time, OUBS brought in quite a few interesting scientific speakers, including at least one Nobel Laureate, though one of the most memorable as Michael Stebbins, who wasn't a researcher and had recently published a book called Sex, Drugs and DNA.

Mary Gregoriou: In my Director of Graduate Studies (DGS) career (~2000-2011) I was very privileged to interact with most academic, teaching, research, administrative and IT staff and research students in the Department of Biochemistry. I was recruited by Professor Raymond Dwek, who had previously introduced a formal graduate skills training programme, to teach and encourage development of professional skills and raise awareness of career choices for graduate employment, as required by Research Councils and other funding bodies.

One day, members of the Biochemical Society committee asked me if I would find out whether the department would help fund a scientific journal written and produced by students and postdocs. I felt that the department would probably say that publishing science is what students and postdocs already do, but I thought it was well worth supporting this request because this was a different opportunity for creativity, less formal, broadening the researchers' interests and understanding of their subject and related areas, well within funding bodies' transferable skills training scope, using teamwork, collaboration and networking skills). I was very pleased that the department supported this student initiative, and perhaps the fact that Phenotype is still an ongoing journal suggests that it is a cherished, enjoyed and worthwhile activity.

Give your cells a **competitive edge**

Sometimes it's the simplest of things that make the difference for cell health.

Anyone who has ever cultured cells in a lab knows that it requires a tremendous investment in time, energy and expense. You just can't leave anything to chance. That's why so many researchers choose Biological Industries (BI) cell culture products. From classic culture media to supplements and reagents, our products are designed to give your cultures superior growth and maintenance performance so you can achieve more consistent and reliable cell health.

Grow a culture of excellence.

No matter what you're growing, we can help you grow it better with an emphasis on quality. Learn more about growing a culture of excellence in your lab.
www.bioind.com

BI
Biological Industries
Culture of Excellence

Biological Industries | T. 972-4-9960595 | info@bioind.com
Biological Industries USA | T. 860.316.2702 | orders@bioindusa.com